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ARTICLES

RADIOACTIVE TAGGING OF *CULEX QUINQUEFASCIATUS* (SAY) WITH P^{32}

R. W. FAY, J. T. BAKER AND J. A. JENSEN [1]

Previous workers (Hassett and Jenkins 1949; Bugher and Taylor 1949; Thurman and Husbands 1951) have demonstrated the value of laboratory determinations of many factors in the rearing, handling, and radioactive tagging of mosquitoes before field dispersion studies are undertaken. Practical consideration must be given (a) to obtaining a suitable initial supply of specimens, (b) to estimating the final adult yield in terms of sex, survival, vigor and radioactivity levels and (c) to determining any species idiosyncrasies in assimilating and retaining the radioactive material. The present paper describes these evaluations as applied to *Culex quinquefasciatus* (Say) adults exposed as larvae to solutions of $Na_2HP^{32}O_4$.

LIFE HISTORY DATA. *Culex quinquefasciatus* (Say), a tropical and subtropical species, is found in foul waters often contained in rain barrels, catch basins, cesspools, ditches, and other similar habitats. The presence of all immature stages, egg rafts, larvae and pupae, in the same breeding sites offers practical difficulties since larvae of uniform age are best suited for tagging with $Na_2HP^{32}O_4$ solutions. Thus, the use of the egg rafts to secure the desired larvae appeared as the best solution. Therefore laboratory studies were made to determine (a) the rate of egg production of adult females, and (b) the possibilities of delaying egg hatch so that eggs from several days could be combined.

Colonies, each containing 100 male and 100 female adults, were offered a chicken as a source of blood every other night. The 5- to 9-day-old females laid an average of 37 egg rafts per colony. In the next 96 hours, an average of 34 egg rafts per colony was obtained. Counts on 38 rafts showed from 110 to 249 eggs per raft with mean and standard deviation values of 158 and 27 eggs per raft, respectively.

At 60°–80° F., essentially 98 percent of the eggs hatched within 24 hours. However, rafts maintained at 50° F. for 24, 48, 72, and 96 hours, showed hatching rates of 89, 97, 72, and 11 percent, respectively, within 24 hours after removal from refrigeration. Thus eggs could be held at 50° F. for 1 to 3 days and combined with freshly laid eggs to secure a uniform starting source of insects.

Eighty-seven of the larvae from approximately 500 eggs per liter of water pupated in 11 to 21 days when reared at 80° F. and 70 percent relative humidity. All successful adult emergence (92.4%) occurred within a 2-day period after pupation. The final distribution of the sexes was essentially equal. The percentages of each sex which emerged from pupae of days 11 through 21 were as follows: for males 8.8, 18.5, 26.9, 14.4, 15.1, 9.4, 2.8, 1.7, 1.0, 1.1, and 0.3, respectively; for females 0.0, 1.9, 6.7, 8.7, 12.2, 29.7, 7.7, 14.5, 9.8, 7.8, and 1.0, respectively.

Based on these production values (namely: 35 egg rafts per 100 females in a 4-day period, a mean number of 158 eggs per raft, 98 percent egg hatch, 87 percent pupation, and 92.4 percent adult emergence with equal numbers of each sex), one million adult females for dispersion studies

[1] From the Communicable Disease Center, Bureau of State Services, Public Health Service, U. S. Department of Health, Education, and Welfare, Savannah, Georgia. A paper from the Proceedings of the 15th annual meeting of the AMCA, Salt Lake City, April, 1959.

would require (a) the collection of 100,000 pupae in a 2-day period and the rearing of the first four days of eggs produced by the resultant adults, or (b) the collection of about 16,000 egg rafts within a 4-day period. The latter starting point appears the more feasible although a combination of methods (a) and (b) could be considered.

To reduce the amount of radioactive solution and the attendant radioactivity hazards, larvae should be concentrated as much as possible during the tagging period. Since crowding might influence the growth and survival values for the species, 3rd instar larvae, reared by the standard insectary procedure, were concentrated at densities of 5,000, 10,000, 15,000, and 20,000 larvae per square foot of water surface. The concentrations were prepared in a series of 8-inch cubical paraffin-lined wooden boxes filled to a depth of 4 inches with tap water (0.5, 1.0, 1.6 and 2.1 larvae per ml. of water, respectively). On the day of concentration, 0.55 to 0.6 mg. of coarsely ground dog biscuit per larvae was provided. During the next 3 days, 0.55, 0.2, and 0.1 mg. of food per larva were added. The pupae were not collected, but daily counts were made on adults caught in cages designed to enclose the wooden boxes.

Each cage consisted of a wooden framework covered on the top and three sides with wire screen and on the remaining side with a cloth sleeve. In the plywood floor of the cage, an opening permitted the cage to slip down snugly over a wooden rearing box. As the cage was lifted off the rearing box, this opening was closed with a sliding panel and simultaneously another panel covered the wooden rearing box. Before removing the cage, all adults were forced from the water surface. Emergence commenced on the 4th day after larval concentration and continued for 5 to 7 days. Adult emergence from the 5,000, 10,000, 15,000 and 20,000 larvae per square foot concentrations were 93, 53, 32 and 8 percent, respectively. At the two lower concentrations, adults were divided equally between the sexes, but at 15,000 larvae per square foot, the females represented only 34 percent of the adults. These tests showed fewer adults from larval concentrations above 10,000 per square foot of water surface. Before these data were fully assembled, however, tests with the actual radioactive material were begun using concentrations of 15,000 larvae per square foot of water surface.

RADIOACTIVE TAGGING DATA. Two series, prepared on successive days using 500 eggs per pan, were reared by routine insectary methods to secure 7-day-old, early 3rd instar and 8-day-old, late 3rd instar larvae, respectively. Larvae were strained from the rearing medium and placed in paraffin-lined wooden boxes at densities of 15,000 larvae per square foot of water surface. Each box, filled to 4 inches, contained 4 liters of water to which 0.1 millicurie $Na_2HP^{32}O_4$ per liter of water was added and thoroughly mixed. The larvae were fed ground dog biscuit according to the schedule described previously. The first adults emerged on the 4th day after concentration of the larvae and emergence continued for 6 to 8 days.

Adults in each cage were collected with a mechanical aspirator and anesthetized lightly with CO_2. Some adults were removed for immediate determination of radioactivity. The remainder were supplied food and held in 1-gallon cardboard cartons for subsequent determinations after 1- and 2-week intervals. For determining radioactivity, each specimen was put into an individual cell, one of a series of holes bored in a wooden paddle and equipped with wire screen tops and bottoms. Specimens of each sex were read in separate paddles.

The mean radioactivity levels for 1-day-old adults from the two age groups of exposed larvae (Table 1) showed highly significant differences between (a) the males and females emerging on the same day from a given set of larvae, and (b) the adults of a given sex emerging the same day from the two sets of larvae. Also, the adults from the exposed 7-day-old larvae

TABLE 1.—Net activity levels (cpm)* of 1-day-old adult *Culex quinquefasciatus* emerging at different intervals after placement of 7- and 8-day-old larvae in solutions containing 0.1 millicurie of $Na_2HP^{32}O_4$ per liter

Days of exposure before emergence	Mean radioactivity (cpm) of adults from larvae exposed at			
	7 days		8 days	
	Male	Female	Male	Female
3	2135	—	290	—
4	1993	2471	419	—
5	1680	2559	539	1050
6	1298	2442	669	1040
7	1105	1924	717	930
8	1020	1801	562	1079
9	847	1485	—	—
10	728	1200	—	—

* Counts per minute.

showed decreasing activity levels as the interval to emergence became longer, whereas the adults from the exposed 8-day-old larvae showed increasing levels (males) or nearly uniform levels (females) as the interval to emergence became longer. Essentially all loss in radioactivity in 8- and 15-day-old adults could be accounted for through decay of the $Na_2HP^{32}O_4$, indicating high retention of absorbed phosphorus by the adults.

Since the adults from larvae exposed at 7 or 8 days of age showed marked differences in radioactivity, further tests were run with 8-, 9-, and 10-day-old larvae in water containing 0.05 millicurie $Na_2HP^{32}O_4$ per liter. Most of the 10-day-old larvae were in the 4th instar. The results (Table 2) re-emphasized the importance of timing the exposure, since the 8-day-old larvae produced adults with activity levels significantly higher than those of adults from larvae exposed at 9 or 10 days. Males from the larvae exposed at 9 days of age showed significantly higher activity levels than males from larvae exposed at 10 days of age. Differences in the adult females were not consistently significant in this latter case. Again with larvae exposed at 8, 9, and 10 days of age,

TABLE 2.—Net activity levels (cpm)* of 1-day-old adult *Culex quinquefasciatus* emerging at different intervals after placement of 8-, 9-, and 10-day-old larvae in solutions containing 0.05 millicurie of $Na_2HP^{32}O_4$ per liter

Days of exposure before emergence	Mean radioactivity (cpm) of adults from larvae exposed at					
	8 days		9 days		10 days	
	Male	Female	Male	Female	Male	Female
3	—	—	491	—	109	—
4	1972	—	736	719	376	500
5	1538	—	527	748	378	563
6	—	—	606	851	508	589
7	—	—	496	800	443	659
8	987	—	466	657	395	584
9	—	—	463	—	287	—
10	638	973	—	477	—	432
11	632	894	—	—	—	—
12	396	798	—	—	—	—

* Counts per minute.

TABLE 3.—Net activity levels (cpm)[a] of adult *Culex quinquefasciatus* from 6- and 7-day-old larvae exposed 24 and 48 hours in water containing 0.05 millicurie of $Na_2HP^{32}O_4$ per Liter

Day of pupation[b]	Day of adult emergence	Mean radioactivity (cpm) of adults from larvae exposed at							
		6 days				7 days			
		For 24-hr.		For 48-hr.		For 24-hr.		For 48-hr.	
		Males	Females	Males	Females	Males	Females	Males	Females
2	4	—	—	—	—	2811	4763	4619	—
	5	—	—	—	—	2469	—	—	—
	6	—	—	—	—	2316	—	—	—
3	4	2959	—	5900	—	2784	5174	5453	8340
	5	2589	—	4774	8242	2446	3903	4867	8933
	6	—	4123	—	—	—	—	4515	—
4	5	2049	3507	4014	8446	1397	3269	—	8318
	6	1088	3065	—	6382	—	3281	—	7416
5	7	—	1609	—	5406	—	—	—	—

[a] Counts per minute.

[b] Day 0 is day of larval exposure to $Na_2HP^{32}O_4$.

the females had significantly higher activity levels than the males; the 8- and 15-day-old adults showed losses in radioactivity almost entirely assignable to normal decay of P^{32}; and the adults from the younger exposed larvae showed progressively lower activity levels as the time to emergence increased, whereas adults from older larvae showed increasing levels with peaks for adults emerging 4 to 6 days after exposure. Although the activity levels for the adult females from 8-day-old larvae exposed to 0.05 and 0.1 millicurie $Na_2HP^{32}O_4$ solutions were quite comparable, parallel observations on the males showed marked differences in the two series (Tables 1 and 2).

To determine the effect of limited 24- or 48-hour exposures on the radioactivity levels of adults, groups of 200 6- and 7-day-old larvae were placed in separate boxes containing 0.05 millicurie $Na_2HP^{32}O_4$ per liter. Half of the larvae from each box were removed at 24 hours, the remainder at 48 hours. Larvae were washed and placed in tap water to complete development. Pupae were collected daily and the early emerging adults for each day's pupae showed the highest activity levels (Table 3). These activity levels were generally higher than those previously obtained, possibly as a result of the differences in numbers of exposed larvae (200 *vs.* 15,000 per square foot). The ratios of activity levels from larvae exposed 48 hours to those from larvae exposed 24 hours were: males—1.89 and females—2.51. This indicates the daily absorption of P^{32} on the 6th, 7th and 8th days was about equal for male larvae but the female larvae absorbed increasing amounts on successive days.

From these studies it appears that the larvae should not be crowded at rates higher than 10,000 per square foot during the exposure period to $Na_2HP^{32}O_4$ and that the length of the exposure may be limited to 48 hours if 6-, 7-, or 8-day-old 3rd instar larvae are utilized. Concentrations of 0.05 and 0.1 millicurie of $Na_2HP^{32}O_4$ per liter did not appear to affect the activity or longevity of the adult mosquitoes.

In view of the somewhat extended period of 7 to 9 days for adult emergence from a given set of exposed larvae, the adults would have to be held if the speed of dispersion was being measured. In this case, adults emerging the first 4 days (principally males) could be destroyed, and adults emerging days 5 to 8 would include about two-thirds of the available females.

SUMMARY. One million radioactively tagged adult female *Culex quinquefasciatus* (Say) for dispersion studies can be obtained (a) by the collection of 100,000 pupae in a 2-day period and the rearing of the first 4 days of eggs produced by the resultant adults, (b) by the collection of about 16,000 egg rafts within a 4-day period and the rearing of the resultant larvae, or (c) by a combination of the two methods (a) and (b). Based on insectary studies, production values are as follows: 35 egg rafts per 100 females in a 4-day period, a mean of 158 eggs per raft, 98 percent egg hatch, 87 percent pupation and 92.4 percent adult emergence, with equal sex division. Exposure of 6- to 8-day-old larvae at concentrations of 10,000 larvae per square foot of water surface for periods of 24 hours or longer in 0.05 to 0.1 millicurie $Na_2HP^{32}O_4$ solutions will yield adults with activity levels detectable for a period of at least two weeks.

ACKNOWLEDGMENT

The authors wish to express their thanks to Mrs. Mary Crawford for her valuable assistance in the determinations of the radioactivity values.

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A MECHANICAL DEVICE FOR SEPARATING THE DEVELOPMENTAL STAGES, SEXES AND SPECIES OF MOSQUITOES[1]

R. W. FAY AND HARVEY B. MORLAN

In large scale production of mosquitoes for investigational purposes, the removal of pupae from the other developmental stages in the rearing containers has often proved a monotonous and time-consuming task. The present paper describes a relatively simple mechanism which performs this operation rapidly and efficiently. Other applications of the apparatus, such as the separation of different species and the separation of the two sexes of a given species are also described.

While the examples noted are confined to the use of the separator in mosquito production, it is felt that wider application may be found for other aquatic organisms in many ecological studies.

APPARATUS. The separator device (Figure 1) consists of a funnel in which the contents of an aquatic insect culture are poured. The culture leaves the funnel through a flattened metal spout and impinging on the upper surface of a base sheet of plate glass (Figure 1, part no. 7), enters a wedge-shaped space between the base sheet and a covering sheet of plate glass (Figure 1, part no. 6). The opening at the upper ends of the glass plates is held constant by metal spacers while at the lower ends of the plates the opening can be precisely regulated with an adjusting screw knob. The lower opening is adjusted so that the larger organisms in the culture are retained between the glass plates while the rest of the culture drains into a receiving container below. Metal guide strips along the sides of the glass plates confine the water and direct its flow into the receiving container. When the retained organisms accumulate in sufficient numbers to impede the water flow, they can be flushed into a separate container by turning the adjusting screw knob to open the lower ends of the glass plates.

RESULTS

GENERAL OPERATION. A suitable adjustment of the separator can be made quite rapidly by visual inspection. With the lower opening almost closed, the various larval instars and pupae of mosquitoes collect in a series of separate bands with the different instars or pupae at various heights between the glass plates and the smallest forms in the lowest band. With gradual increase of the lower opening, the smaller organisms are flushed out and the bands of larger organisms descend between the glass plates until the desired stage represents the lowest band. If the stage to be separated is not the largest, *e.g.*, the removal of the 4th-instar larvae from younger larvae and pupae, the first adjustment releases the young larvae and a second adjustment releases the 4th-instar larvae while the pupae are retained in the separator. Once the desired settings have been determined, calibrations on the adjustment screw knob permit rapid duplication of any given setting. With 16 divisions on the knob, each division changes the width of the lower opening by 0.0014 inch on the model described.

SEPARATION OF MOSQUITO PUPAE. In removing pupae from mixed culture of larvae and pupae, two factors have influenced the speed of operations: (a) the frequency with which flushing is necessary and (b) the rate of water flow through the separator. From 800 to 900 pupae can be collected in the separator before large larvae are blocked and flushing becomes necessary. At settings required to retain mosquito pupae, one liter of water takes about 45 seconds to pass through the separator. In

[1] From the Communicable Disease Center, Bureau of State Services, Public Health Service, U.S. Department of Health, Education, and Welfare, Savannah, Georgia.

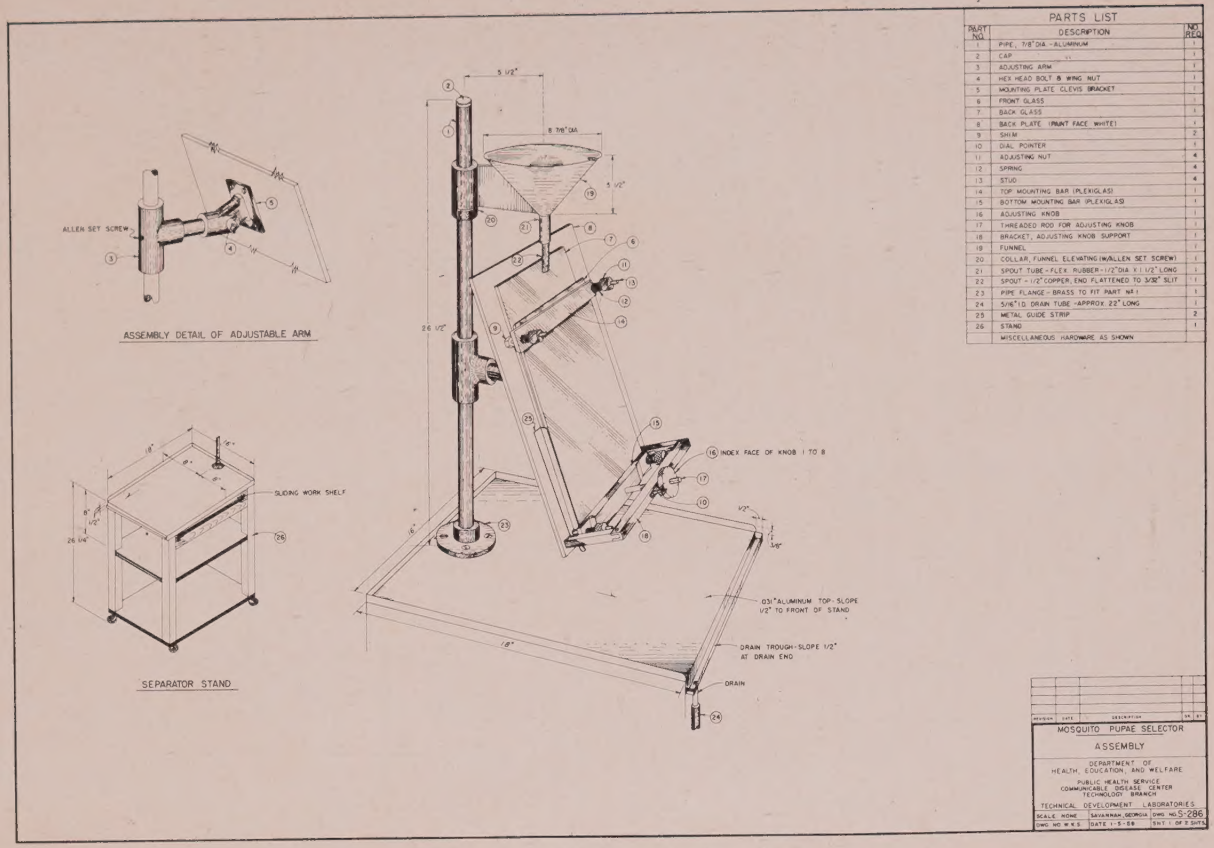
PARTS LIST
PART NO. | DESCRIPTION | NO. REQ.
1 | PIPE, 7/8" DIA - ALUMINUM | 1
2 | CAP " " | 1
3 | ADJUSTING ARM | 1
4 | HEX HEAD BOLT & WING NUT | 1
5 | MOUNTING PLATE CLEVIS BRACKET | 1
6 | FRONT GLASS | 1
7 | BACK GLASS | 1
8 | BACK PLATE (PAINT FACE WHITE) | 1
9 | SHIM | 2
10 | DIAL POINTER | 1
11 | ADJUSTING NUT | 4
12 | SPRING | 4
13 | STUD | 4
14 | TOP MOUNTING BAR (PLEXIGLAS) | 1
15 | BOTTOM MOUNTING BAR (PLEXIGLAS) | 1
16 | ADJUSTING KNOB | 1
17 | THREADED ROD FOR ADJUSTING KNOB | 1
18 | BRACKET, ADJUSTING KNOB SUPPORT | 1
19 | FUNNEL | 1
20 | COLLAR, FUNNEL ELEVATING (WALLEN SET SCREW) | 1
21 | SPOUT TUBE - FLEX RUBBER - 1/2" DIA X 1 1/2" LONG | 1
22 | SPOUT - 1/2" COPPER, END FLATTENED TO 3/32" SLIT | 1
23 | PIPE FLANGE - BRASS TO FIT PART NO 1 | 1
24 | 5/16" I.D. DRAIN TUBE - APPROX. 22" LONG | 1
25 | METAL GUIDE STRIP | 2
26 | STAND | 1
 | MISCELLANEOUS HARDWARE AS SHOWN
ALLEN SET SCREW
ASSEMBLY DETAIL OF ADJUSTABLE ARM
SLIDING WORK SHELF
SEPARATOR STAND
5 1/2"
8 7/8" DIA
5 1/2"
26 1/2"
16 INDEX FACE OF KNOB 1 TO 8
.031" ALUMINUM TOP - SLOPE 1/2" TO FRONT OF STAND
18"
DRAIN TROUGH - SLOPE 1/2" AT DRAIN END
DRAIN
MOSQUITO PUPAE SELECTOR
ASSEMBLY
DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
PUBLIC HEALTH SERVICE
COMMUNICABLE DISEASE CENTER
TECHNOLOGY BRANCH
TECHNICAL DEVELOPMENT LABORATORIES
SCALE NONE
SAVANNAH, GEORGIA
DWG. NO. S-286
DWG NO M K S
DATE 1-5-58
SHT 1 OF 2 SHTS

FIG 1.—Mosquito pupae selector.

ordinary insectary production with cultures ranging from 5 to 100 pupae per liter of water, the separation of 1000 pupae requires about 20 minutes. In contrast, the picking of pupae from similar cultures with small screen-wire spoons by skilled operators requires about 90 minutes per thousand pupae.

Tests have been made to determine any possible injury to the pupae processed through the separator. Emergence and survival rates of adult mosquitoes from pupae collected with the separator and those collected with the screen-wire spoon technique showed no significant differences.

SEPARATION OF MOSQUITO SPECIES. In field collections of mosquito larvae and pupae, other invertebrate predators or competitors often are taken inadvertently. The separator has been useful in rapidly excluding most of these forms where a size differential existed between the organisms and the desired mosquito specimens.

In certain instances, the separator has been employed to divide mixed cultures of two mosquito species into fractions containing essentially only a single species. A mixed culture of *Aedes sollicitans* and *A. taeniorhynchus* pupae were processed through the separator by widening the lower opening stepwise and flushing the separator between adjustments to remove the pupae in successive size groups. Each size group was then caged and the emerging adults identified (Table 1). Over a 14-day period, the 4500 pupae studied showed a distribution by species and sex of 34.5 percent male and 38.3 percent female *A. taeniorhynchus* and 13.7 percent male and 13.5 percent female *A. sollicitans*. In separating the two species, the separator could be adjusted to a slit width of 0.057 inch and the pupae which passed through the slit would comprise at least 94 percent *A. taeniorhynchus*. This sample would contain 97 and 88 percent of the available male and female *A. taeniorhynchus* pupae. The separator could be adjusted then to an opening of 0.060 inch, and the next sample flushed would contain 2 and 11 percent of the total male and female *A. taeniorhynchus* pupae mixed with 22 and 16 percent of the total male and female *A. sollicitans* pupae. The separator could be opened fully and the remaining pupae would consist of at least 96 percent *A. sollicitans*. This sample would have 70 and 78 percent of the available male and female pupae of *A. sollicitans*.

TABLE 1.—Partial separation of a mixed culture [a] of *Aedes sollicitans* and *Aedes taeniorhynchus* pupae by successive settings of the pupae separator

Width of opening	Species composition (percent) of sample passing opening				Cumulative percent of population removed			
	A. *taen.*		A. *soll.*		A. *taen.*		A. *soll.*	
	M	F	M	F	M	F	M	F
.045"	90	10	—	—	4	0.5	—	—
.048"	78.5	20	—	1.5	13.5	2.5	—	0.5
.051"	71	28	0.5	0.5	50	15	0.5	1.0
.054"	46	53	0.5	0.5	83	50	2.0	2.5
.057"	24	70	4	2	97.5	88	8.0	6.0
.060"	9	40	30	21	99	99	30	22
.062"	1	3	54	42	99.5	99.5	66	50
.065"	0.5	1	47	51.5	100	100	88	74
.068"	—	—	35	65	—	—	98.5	94.5
.071"	—	—	23	77	—	—	100	99
.074"	—	—	—	100	—	—	—	100

[a] Culture composition 34.5 percent male and 38.3 percent female A. *taeniorhynchus* and 13.7 percent male and 13.5 percent female A. *sollicitans*.

SEPARATION OF MOSQUITO SEXES. In certain research work the investigator may have interest in only one sex of the adult insects. The separator has been employed in the sex separation of mosquito pupae with greater or less degrees of success depending upon the species involved. The results of sex separation of the pupae from insectary populations having essentially equal numbers of each sex are shown in Table 2. The effectiveness of the separation can be based on two values: (a) the percentage of the desired sex in a sample obtained with a given slit setting, and (b) the percentage of the total available members of a given sex contained in the sample. For example, the male sample of *Aedes taeniorhynchus* obtained through a slit opening of 0.045 inch (Table 2) contained 90 percent males mixed with 10 percent females. These males represented 85 percent of all the males in the population under separation. Based on these factors, the species tested are listed in order of increasing effectiveness of the separator in dividing the sexes of the species. The best sex separation occurs in those species with the smallest pupae and the most rapid development.

TABLE 2.—Partial separation of male and female pupae of various species of mosquitoes by successive settings of the pupa separator

Species	Sex sample desired	Slit width (inch)	Sex division: % in sample		Sex division: % of total culture	
			Male	Female	Male	Female
Aedes sollicitans	Male	.060	59	41	31	22
	Mixed	.065	53	47	57	52
	Female	.065+	33	67	12	26
Anopheles quadrimaculatus	Male	.047	77	23	55	11
	Mixed	.049	36	64	35	42
	Female	.049+	12	88	10	47
Aedes taeniorhynchus	Male	.045	90	10	85	10
	Mixed	.047	32	68	13	28
	Female	.047+	3	97	2	62
Culex quinquefasciatus	Male	.039	95	5	91	9
	Mixed	.042	59	41	8	13
	Female	.042+	10	90	1	78
Aedes aegypti	Male	.040	98	2	87	2
	Mixed	.043	44	56	10	15
	Female	.043+	5	95	3	83

SUMMARY. A mechanical device is described which permits the rapid and accurate separation of mosquito pupae from the larvae in insectary culture containers. The separator has also been useful in separating species commonly found together in field collections such as *Aedes sollicitans* and *Aedes taeniorhynchus*. Further applications of the separator have been found in dividing the pupae of certain mosquito species into male, mixed, and female subsamples.

OBSERVATIONS ON THE SALINITY ON NATURAL AND IMPOUNDED SALT-MARSH AREAS IN NEW JERSEY IN RELATION TO VEGETATION, MOSQUITOES AND PREDATORS

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The mosquito-breeding potential of existing and proposed impoundments and adjacent unimpounded salt-marsh areas in New Jersey was studied in cooperation with the State Division of Fish and Game and Agricultural Experiment Station from 1953 through 1955.[1] Descriptions of the areas, the techniques used, and the results of the study were given by Chapman and Ferrigno (1956). This paper covers observations made on salinity in relation to vegetation, mosquitoes, and their predators not heretofore reported.

Horsfall (1955) presented a general compilation of the reported salinity tolerances of many mosquito species but the paucity of the data is quite evident. Darsie and Springer (1957), reporting on a companion study of some Delaware impoundments and salt-marsh areas, mentioned some of the misconceptions that have accrued with respect to the salinity tolerance and breeding habits of *Culex salinarius* Coq. Many of them exist because insufficient observations have been made in different habitats. Even Carpenter and La Casse (1955) make no mention of this species breeding in saline waters.

Salinity data were taken monthly from one station in each vegetative or ecological type in six fresh-water impoundments in 1954 and 1955 and from four unimpounded salt-marsh areas in 1953 through 1955 (Table 1). Simultaneous observations were made on the presence and identity of mosquito larvae (Table 2) and potential predators (Table 3). Salinities were based on the total chloride as determined by the reaction of the water sample with silver nitrate and a color indicator and are expressed as the percent of the mean salinity of the Atlantic Ocean, which is 32,210 p.p.m. The "number of readings" column in the tables indicates the number of times that a plant, mosquito, or predator occurred in the sampling program. The presence of *Anopheles, Culex,* and especially *Aedes* larvae, in the vicinity of a plant in water of a known salinity does not necessarily indicate a site of oviposition, but only denotes the collection of the larvae in that locality. The concentration of receding waters containing mosquito larvae into potholes and depressions is a phenomenon well understood but often overlooked.

As shown in Table 1, the highest mean salinities in the impounded areas were tolerated by common three-square, soft stem bulrush; bare areas, Olney's three-square, and common reed; the lowest mean salinities occurred in stations along the wood line and adjacent to it which contained sedge. The highest single reading was 44.6, noted in Olney's three-square. In the unimpounded areas the highest mean readings were observed in salt-marsh cord grass, black grass, and salt-meadow cord grass, and the lowest in saltgrass and switch-grass along the upland margins. The highest readings were 137.0 and 115.5 in salt-marsh cord grass and 101.8 in black grass.

The previous report (Chapman and Ferrigno) showed that in the impoundments cattail, common reed, and soft-stem bulrush in that order always produced the

[1] Acknowledgment is made to the New Jersey Division of Fish and Game, especially L. G. MacNamara, for the unlimited use of its facilities at the Tuckahoe Public Shooting Grounds, and also to F. Ferrigno for assistance in the collection of the field data and acquainting the writer with the flora of the study areas.

TABLE 1. Range of salinity in the immediate vicinity of plant species or ecological types in the New Jersey impounded and unimpounded salt-marsh areas, 1953–55, expressed as percent of mean ocean salinity

Plant or ecological type	Impounded areas			Unimpounded areas		
	Number of readings	Range	Mean	Number of readings	Range	Mean
Bare	6	0.9–18.5	9.3	—	—	—
Carex sp. (sedge)	7	.3– 1.5	0.7	—	—	—
Distichlis spicata (L.) (saltgrass)	—	—	—	15	7.4– 45.6	25.5
Ditches	9	.3–13.7	2.6	12	11.0– 83.6	47.9
Eleocharis olivacea Torr. (spike rush)	7	1.5– 5.1	2.6	—	—	—
Juncus gerhardi Loisel (black grass)	—	—	—	30	5.9–101.8	50.9
effusus L. (soft rush)	33	.3–35.3	3.5	—	—	—
Panicum virgatum L. (switch grass)	18	.6–39.2	6.1	32	1.5– 76.9	20.5
Phragmites communis Trin. (common reed)	19	.8–35.2	8.6	—	—	—
Scirpus americanus Pers. (common three-square)	11	.9–36.5	11.5	—	—	—
olneyi Gray (Olney's three-square)	39	.3–44.6	8.7	56	4.0– 83.0	35.5
validus Vahl. (soft-stem bulrush)	7	1.7–30.8	10.8	—	—	—
Typha angustifolia L. and *latifolia* L. (cattail)	34	.5–43.2	7.5	—	—	—
Spartina alterniflora (Loisel) (salt-marsh cord grass)	—	—	—	61	8.8–137.0	52.3
patens (Ait.) (salt-meadow cord grass)	—	—	—	56	1.8– 97.3	40.9
alterniflora and *patens* mixture	—	—	—	17	12.1– 55.8	27.9
Wood line	7	.3– .5	.4	—	—	—

largest numbers of *Culex salinarius*. All these plant species were found in areas with high mean salinities. Also it was shown that bare areas and those with soft-stem bulrush, both with high mean salinities, gave the highest counts of *Anopheles bradleyi* King in the impoundments.

As shown in Table 2, *Aedes sollicitans, Aedes cantator, Anopheles bradleyi,* and *Culex salinarius,* generally known as salt-marsh mosquitoes (Headlee 1945), tolerated both the highest maximum and mean salinities in both the impounded and unimpounded areas. *Aedes canadensis, Mansonia perturbans,* and *Anopheles crucians* appeared to be the least tolerant of all species to high salinities. *M. perturbans* was generally found abundant only in impoundments that remained flooded and these flooded impoundments were always the least saline. Drawing down impoundments usually prevented their breeding (Chapman and Ferrigno).

In data not shown, an impoundment in 1953 with salinities up to 12.0 percent produced fair numbers of *Anopheles quadrimaculatus,* but none were found the next 2 years, when much higher salinities were recorded.

The maximum salinities in which the various species were found breeding were as follows: *Aedes sollicitans* 137.0, *Aedes cantator* 109.4 and 98.2, *Culex salinarius* 83.6, and *Anopheles bradleyi* 55.8. Since the data show that within the impoundments *Culex salinarius* was consistently dipped from water with a high mean salinity, and that this was the most abundant species in cattail, common reed, and soft-stem bulrush, all of which were in areas with high mean salinities, it is obvious that it prefers saline conditions to fresh water in this general area.

The most tolerant predators in the impoundments appeared to be a giant water bug (*Belostomatidae*), predacious diving

TABLE 2.—Range of salinity in New Jersey impounded and unimpounded salt-marsh areas in which various mosquito larvae were dipped, 1953–55, expressed as percent of mean ocean salinity

Mosquito species	Impounded areas			Unimpounded areas		
	Number of readings	Range	Mean	Number of readings	Range	Mean
Aedes canadensis (Theob.)	13	0.3– 4.8	1.2	1	—	15.1
cantator (Coq.)	45	.3–44.6	7.4	49	1.5–109.4	40.0
sollicitans (Wlkr.)	35	.9–44.6	11.0	58	11.0–137.0	49.5
vexans (Meig.)	24	.3–33.3	5.5	4	1.5– 27.5	9.4
Anopheles bradleyi King	18	.5–44.6	10.0	50	4.6– 55.8	28.0
crucians Wied.	18	.3–16.7	4.3	—	—	—
Culex restuans Theob.	5	.9–17.5	6.0	—	—	—
salinarius Coq.	61	.5–43.2	8.9	25	11.0– 83.6	27.4
territans Wlkr.	33	.3–33.3	4.6	—	—	—
Culiseta inornata (Will.)	—	—	—	1	—	24.5
Mansonia perturbans (Wlkr.)	7	.5– 2.4	1.6	—	—	—

TABLE 3.—Range of salinity in New Jersey impounded and unimpounded salt-marsh areas in which various predators of mosquito larvae and pupae were observed, 1953–55, expressed as percent of mean ocean salinity

Predator	Impounded areas			Unimpounded areas		
	Number of readings	Range	Mean	Number of readings	Range	Mean
Coleoptera						
Dytiscidae (predacious diving beetles)	70	0.3–44.6	7.6	30	1.5– 67.0	24.6
Hydrophilidae (water scavenger beetles)	2	.5– 1.4	1.0	4	18.5– 72.7	39.9
Heteroptera						
Belostomatidae						
Belostoma lutarium (Stol.)	13	.8–43.8	9.6	9	6.8– 32.5	16.2
Naucoridae						
Pelocoris femoratus (P. de B.)	7	.8–26.1	6.7	—	—	—
Notonectidae						
Notonecta spp.	8	.8– 5.9	3.4	—	—	—
Odonata						
Damselfly naiads	18	.4–14.2	3.4	3	6.2– 12.1	8.5
Dragonfly naiads	12	.3–14.2	4.5	22	4.1– 92.8	33.1
Minnows	19	.3–35.3	6.2	130	6.8–109.4	39.4

beetles, a creeping water bug (*Naucoridae*), and minnows. Water scavenger beetles, minnows (principally *Cyprinodon variegatus* Lace. and *Fundulus* spp.), and dragonfly naiads were the most tolerant species in the unimpounded areas. High salinities were no barrier to some of these predators, since minnows and dragonfly naiads were noted in waters containing salinities of 109.4 and 92.8 percent, respectively.

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THE INFLUENCE OF SOIL FERMENTATION ON OVIPOSITION SITE SELECTION BY MOSQUITOES

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Studies of the natural history of mosquitoes show that each species has rather characteristic breeding habits. The habits of the family as a whole are sufficiently diverse that the larvae of one or more species of mosquitoes are able to inhabit almost every type of terrestrial water accumulation.

Field observations indicate clearly that the ecological distribution of larvae is strongly influenced by the oviposition habits of the adults of different species. While the degree of habitat specificity of mosquito species is variable there is no evidence that adults deposit their eggs in all types of water, with observed distribution resulting from a selective mortality governed by the relative compatibility with the particular environment thus chosen at random.

Laboratory experiments on oviposition behavior have, in general, given unsatisfactory results. The reactions of mosquitoes in the restricted environment of a cage often seen quite unrelated to the reactions that we know or suspect exist in nature. In the laboratory, mosquitoes often will oviposit in anything that contains water, even solutions lethal to eggs or larvae. When mosquitoes have evidenced discrimination toward a series of oviposition media, the basis of selection has been difficult to evaluate.

Many authors have dealt with the influence of inorganic compounds dissolved in breeding waters on oviposition site selection. This work has been adequately reviewed by other authors, particularly Wallis (1954) and Hudson (1956) and will not be repeated here.

Few investigators have considered the possibility that organic compounds attract adult mosquitoes to oviposition sites. Rudolfs (1922), in a study of the chemotropisms of mosquitoes, demonstrated that adult female mosquitoes are attracted through olfactory perception toward several compounds.

Rudolfs and Lackey (1929) studied two pools that differed greatly in production of mosquitoes. Mosquito breeding could not be correlated with pH, free carbon dioxide, chlorides, sulfates, acid carbonates, free ammonia, or albuminoid ammonia. However, a marked difference was noted in the decomposition products of the mixtures of muck and organic material obtained from the bottoms of the pools. It appeared that selection of breeding site might have been influenced by the presence of specific organic substances in the water produced from the decomposition of organic matter.

Observations. In California, the most important species in vector control work is *Culex tarsalis* Coquillett, the principal vector of encephalitis. This ubiquitous species commonly breeds in rice fields, a well-defined environment.

The amounts of organic material in the soil of rice fields are moderate to large, depending on the agricultural practice followed between rice crops, during the winter. Some growers disk the rice straw into the soil in the fall, some burn (incompletely) and disk in the spring, others grow vetch over the rice stubble and disk all the plant material into the soil in the spring. Field observations indicate that increased mosquito breeding is associated with the latter practice, which places the greatest amount of plant material in the soil at the time the fields are flooded.

In well-aerated soils plant material is normally decomposed through oxidation by aerobic bacteria and fungi. In contrast, submergence of the soil permits only anaerobic bacterial decomposition of the plant material, producing butyric acid, propionic acid, lactic acid, acetic acid,

methane, hydrogen, and carbon dioxide. This fact is well known, and was shown by Acharya (1935 a and b) to be specifically true of rice straw.

These reduced organic compounds diffuse into the overlying water where their fate is determined by a number of factors. All are subject to oxidation, ultimately to carbon dioxide, by aerobic bacteria in the surface water. Hydrogen and methane escape readily into the atmosphere. Acetic acid and butyric acid, although soluble in water, exert sufficient vapor pressures that considerable proportions are liberated into the air. Carbon dioxide remains in the water, partly as carbonates, and is available for assimilation by algae.

In 1956, the author treated experimental rice field plots with specific chemical algicides. Certain treatments markedly depressed algal growth with a resulting reduction in the biochemical oxygen demand (BOD) of the water. The appreciably reduced pH of the water probably resulted from the accumulation of organic acids. The lower BOD values most probably indicated a lag in oxidation of the organic acids produced from the underlying soil. Oviposition by *C. tarsalis* was greatly favored in the algicide-treated plots. The observations suggest that, because of inefficient oxidation in the water, some product of anaerobic decomposition was accumulating and serving as an attractant for the adult female mosquitoes. The basis for choice was evidently a preference for algicide-treated plots (Gerhardt, 1957).

The unanswered questions posed by these observations show the need for investigation of the influence of decomposition of organic material in rice field soil on preferential selection of oviposition sites.

First Experiment

Methods. In July, 1957, at a location near Davis, California, 13 small plots were constructed by digging shallow depressions two-feet square and lining them with polyethylene sheeting. The plots were in 2 rows about 3 feet apart, with those in each row separated by one foot of soil

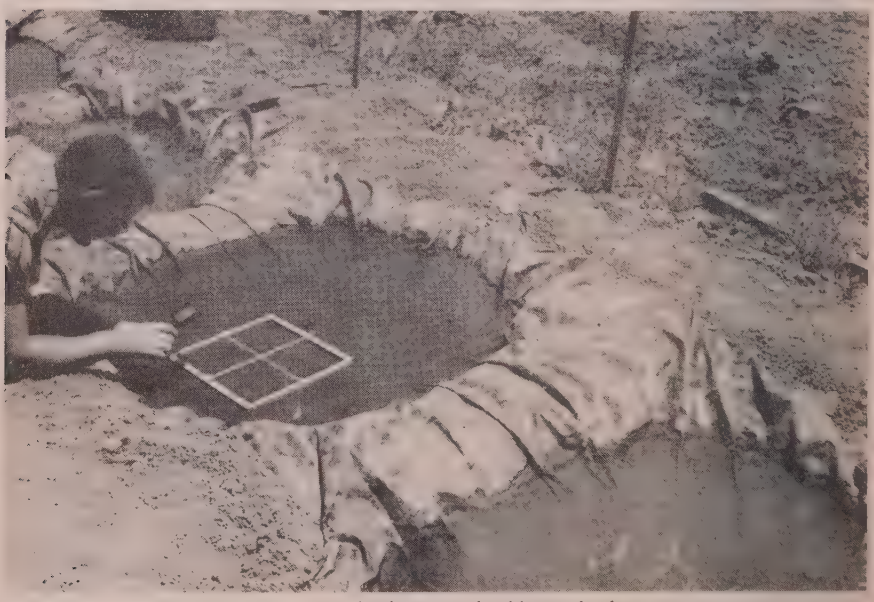

Fig. 1.—Counting larvae—oviposition study plots

covered with the continuous polyethylene sheet. A 6-inch layer of Stockton clay adobe soil (a common California rice soil) was placed on the sheeting in the plots.

For extra organic matter, 5 of the 13 plots each received 2 pounds of a prepared dog meal (gross composition listed as 25 percent protein, 6 percent fat, 5 percent fiber, 10 percent ash). The meal was supplied to the soil below the depth to which reducing conditions would extend.

The plots were flooded with tap water to a depth of six inches, and the depth was restored daily. No circulation was provided, and the only water loss was through evaporation.

The progress of decomposition of the organic material supplied to the plot soils was closely followed. The accumulation of organic acids in the surface water was determined by periodic pH measurements and by chemical analysis of the water for volatile acids. The development of reducing conditions in the soil was determined with a platinum electrode.

Oviposition preference was measured by counting egg rafts deposited on the plot surfaces and by counting first instar larvae only. Larvae were calculated from counts of those enclosed in a rack one foot square, floating on the water (Figure 1).

Results of First Experiment. Egg rafts, first observed in the organically fortified plots six days after flooding, were left in place. The resulting larvae were later identified as *Culex peus* (= *stigmatosoma* Dyar). Determination of pH of the water and reduction-oxidation potentials of the soil showed that reducing conditions were well established in the treated plots, with acids accumulating in the surface water.

Oviposition continued throughout several weeks. Larvae of *C. peus* predominated throughout the test period, but, during the last week, specimens of *C. tarsalis* were abundant and several specimens of *Anopheles freeborni* Aitken were recovered. The control plots remained completely free of egg rafts and larvae throughout the test period. A summary of the results is presented in Table 1.

Second Experiment

Methods. Following the first experiment, all of the plots were drained, cleaned, and flushed out with fresh water. The major portion of the added organic material had been fermented; the rapid evolution of gases that characterized the early stages of the experiment had stopped.

Three plots previously used as controls were chosen for a supplementary experiment, as follows: (a) one pound of sucrose in strong solution was injected into the soil of one plot; (b) the same amount of casein solution was injected into the soil of the second plot; (c) the third plot received one pound of hydrogenated vegetable oil.

Results of Second Experiment. Five days after these organic materials were introduced into the three test plots egg rafts were found on the water surface of all test plots. None were observed on the control plots. The resulting larvae proved to be *Culex peus*.

Rafts continued to appear in the sucrose plot for 15 days after they were first found. The casein and fat plots continued to attract ovipositing female mosquitoes for 24 and 31 days, respectively, after the first rafts were found.

TABLE 1.—Main oviposition study. Data collected from June 6, 1957, through July 12, 1957

	Total no. egg rafts	Average no 1st instar larvae/sq. ft.	Average pH water	Volatile acids mg/liter
Test plots (5) organic material added to soil	20	123	8.06	54.2
Control plots (8)	0	0	8.70	30.7

A summary of the results is presented in Table 2.

TABLE 2.—Supplementary oviposition study

	No. rafts recovered	Av. pH during period of oviposition	Av. pH after oviposition ceased	Period of active oviposition (days)
Sucrose added	10	8.1	9.0	15
Casein added	14	8.4	9.0	24
Fat added	16	8.5	8.8	31
Control	0	8.9	8.9	(observed for 35 days)

DISCUSSION. The foregoing experiments suggest that adult female mosquitoes were attracted to the test plots by compounds resulting from the anaerobic decay of organic materials. A relatively simple, easily fermentable substance (sugar) produced a brief response by the mosquitoes; more complex materials (casein and fat) produced longer responses.

Eggs were first deposited on the water on the day when the pH values started to drop. In the plots studied, this increase in acidity can be attributed to the accumulation of organic acids from fermentation of the organic materials supplied to the plots.

A shallow-water environment includes two distinct components of vastly different characteristics. The water-logged soil becomes anaerobic shortly after being flooded, while the soil-water interface and the overlying water remain aerobic, increasingly so as photosynthetic organisms become established. The metabolism of the main body of soil is fermentative, and that of the water is oxidative. Under ordinary circumstances, the two functions are approximately balanced.

The environment can be artificially altered so that, for a time, the fermentative metabolism is faster than the oxidative metabolism. If extra organic material is supplied to the soil, fermentation products accumulate in the water. Similarly, when the oxidative biota of the water is reduced—by such artificial means as treatment with copper sulfate or other algicidal materials—the fermentation products accumulate.

With adequate knowledge of the particular environment, it is possible to judge the nature of its predominant metabolism as excessively fermentative, approximately balanced, or predominantly oxidative. These three characteristics have long been recognized by limnologists in regard to larger bodies of water. The oxidative environment is designated as oligotrophic, the balanced environment as eutrophic, and the fermentative environment as dystrophic.

The writer prefers not to borrow the exact terms of the limnologist to describe the trophic relationships of shallow bodies of water. The condition of dystrophy in lakes is intense, hardly comparing with the condition found in shallow-water environments.

If the limited data of the experiments are to be interpreted precisely, the appearance of *C. peus* in the study plots during the period of rapid fermentation confirms the preference of this species for foul water. The succession of *C. tarsalis* and *A. freeborni* in apparent response to reduced fermentative activity, conforms to the oviposition preferences of these species. However, these observations are in need of exhaustive investigation.

Apparently *Culex peus* prefers environments that display the characteristics of the fermentative, anaerobic soil. The exclusive manner in which the test plots were selected suggests that these characteristics were detected from a considerable distance. It would seem that the ovipositing females were attracted by olfactory stimuli directly to the plots that had extra organic matter in the soil.

The exact nature of the attractants re-

mains unknown, but the short-chain volatile acids known to arise from water-logged soils seem, logically, to be the compounds most directly associated with the attraction of the mosquitoes.

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A LIST OF NEVADA MOSQUITOES, WITH FIVE NEW RECORDS

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Some of the first records of mosquitoes collected in Nevada were by Dyar (1917, 1922), who reported 8 species from the State. Carpenter and LaCasse (1955) listed 12 species from Nevada. The most recent list by Richards *et al.* (1956), contains 23 species and is based on their published records plus their personal collections.

While surveying areas in the counties surrounding Reno, the writer collected larvae of four species not heretofore reported from the State. An additional species was identified from larval material collected by R. C. Bechtel, of the Nevada State Department of Agriculture. These records are as follows:

Aedes communis (DeGeer): Douglas County: Spooner Summit, III-19–59; Glenbrook, IV-24–59. Washoe County: Lake Tahoe, IV-9–59; Mt. Rose, IV-21–59.

A. hexodontus Dyar: Douglas County: Spooner Summit, III-19–59; Glenbrook, IV-24–59. Ormsby County: Lake Tahoe, IV-9–59. Washoe County: Mt. Rose, IV-21–59. Elko County: Lamoille Canyon, VI-2–59.

A. pullatus (Coq.): Elko County: Lamoille Canyon, VI-18–58 (collected by R. C. Bechtel), VI-2–59.

A. schizopinax Dyar: Washoe County: Mt. Rose, IV-21–59; Hunter Lake, VI-11–59.

Culex territans Walker: Douglas County: Lake Tahoe, IX-1–58. Ormsby County: Lake Tahoe, X-10–58. Washoe County: Lake Tahoe, VII-4–59.

A record apparently overlooked by both Carpenter & LaCasse and Richards *et al.* is that of an *Anopheles* reported by Dyar (1922) as *maculipennis* Meigen from Steamboat Springs, VIII-19–15. The wing tips of the species known by Dyar as *maculipennis* have a brassy reflection and must be either *occidentalis* Dyar and Knab or *earlei* Vargas. The former species is supposedly restricted to a rather narrow strip along the Pacific coast from California to Canada; hence the Nevada record probably is *earlei*.

[1] In cooperation with the Nevada Agricultural Experiment Station.

TABLE 1.—List of mosquitoes recorded from Nevada

Species	Authority
Aedes campestris Dyar and Knab	Richards *et al.* 1956
cataphylla Dyar	Richards *et al.* 1956
cinereus Meigen	Richards *et al.* 1956
communis (DeGeer)	
dorsalis (Meigen)	Dyar 1922
fitchii (Felt and Young)	Richards *et al.* 1956
hexodontus Dyar	
idahoensis (Theobald)	Dyar 1922
increpitus Dyar	Dyar 1922
melanimon Dyar	Carpenter and LaCasse 1955
nigromaculis (Ludlow)	Richards *et al.* 1956
niphadopsis Dyar and Knab	Carpenter and LaCasse 1955
pullatus (Coq.)	
schizopinax Dyar	
sierrensis (Ludlow)	Dyar 1922
vexans (Meigen)	Richards *et al.* 1956
Anopheles earlei Vargas	Dyar 1922
franciscanus McCracken	Carpenter and LaCasse 1955
freeborni Aitken	Carpenter and LaCasse 1955
Culiseta impatiens (Walker)	Richards *et al.* 1956
incidens (Thomson)	Dyar 1922
inornata (Williston)	Dyar 1922
Culex boharti Brookman and Reeves	Richards *et al.* 1956
erythrothorax Dyar	Richards *et al.* 1956
peus Speiser	Richards *et al.* 1956
tarsalis Coq.	Dyar 1922
territans Walker	
thriambus Dyar	Richards *et al.* 1956
Uranotaenia anhydor Dyar	Carpenter and LaCasse 1955

The 29 species now recorded from Nevada are listed in Table 1. Approximately 10 additional species have been reported from adjacent counties of California, Arizona, and Utah.

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CULISETA MELANURA (COQUILLETT) AND EASTERN EQUINE ENCEPHALITIS IN CONNECTICUT

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Culiseta melanura is a mosquito which has become the subject of special investigation since 1951, when the virus of eastern equine encephalitis (EEE) was isolated from it by Chamberlain, *et al.* (1). This mosquito occurred at the site of an encephalitis epidemic among penned pheasants at Farmington, Connecticut, in 1953, and observations on its biology were reported by Wallis (2). During the same year isolations of EEE virus were reported by Holden, Miller and Jobbins (3) from three pools of *C. melanura* collected in New Jersey and intensive study of its biology was initiated in that state. Since then studies of virus-vector relationships indicated that *C. melanura* was a good host and was capable of transmitting the EEE virus in the laboratory (Chamberlain *et al.* (4)). In 1955, observations were reported by Chamberlain, Sudia and Nelson (5) on the biology of this species in the laboratory and during the following year Burbutis and Lake (6) reported bionomic field studies in New Jersey. During the 1956 season, four isolations of virus were obtained from pools of *C. melanura* in New Jersey (Chamberlain *et al.* (7)), and five virus isolations were obtained from pools of this species in Massachusetts (Feemster *et al.* (8)). It began to look as if *C. melanura* might be a primary vector of transmission of the EEE virus. However, our studies in Connecticut between 1953 and 1958 have produced very little evidence in the field to support this hypothesis.

EEE in Connecticut has been an agricultural and wildlife problem since there has been no confirmed human case of the disease (Jungherr and Wallis (9)). Between 1938 and 1958, presence of virus has been confirmed on 25 farms in Connecticut either in horses or pheasants. The mosquito population of 20 of these farms was investigated at the time of virus activity. From these studies 15,819 adult female mosquitoes were collected and identified. Of these, 1,730 were *C. melanura,* most of which were collected at Farmington, Connecticut. This was the only one of the 25 sites where *C. melanura* was found repeatedly. It was encountered only once at two other of the 25 sites, and was not collected in the other 22 areas where virus activity has occurred. Therefore, this species must be considered rare in Connecticut, and the correlation of its distribution with that of the incidence of the disease has not been confirmed.

While it was not possible to collect this species from all epidemic sites, the one area where it could be found readily provided bountiful material for studies of its ecology. Many of the details reported by Burbutis and Lake (6), such as the spring and fall seasonal peaks in the adult population, and the overwintering of larvae were observed in the field. It was particularly interesting to find that during the 1957 and 1958 seasons, most of the blood feeding of the adult females occurs during the June population peak. At this time, as high as 50 percent of the adult females were either freshly blooded or gravid with eggs. Particular efforts were made in collecting when bird nesting was at its peak. It was disappointing to find that even though nesting birds in an area contained EEE antibody, and a high proportion of the *C. melanura* population were engorged with avian blood, there was no virus isolation from pools of this species (Taylor (10)).

At the Farmington study site, conditions were ideal for investigation of the feeding activity of the mosquito population as it related to both wild and domestic birds. Each year, early in July, 400 young pheasant chicks were obtained by a local hunt club and released in a large open range pen

where they were reared to adult pheasants. Within 100 yards of the pen there were diurnal resting places which provided excellent sites for sampling the *Culiseta* population in the area. The numbers of female mosquitoes in the routine collections which contained fresh avian blood meals were recorded for comparison of the percentage of fed specimens prior to and after the time when pheasant chicks were introduced into the environment. During the early period, when only wild bird nestlings were available for feeding, it was found that from 3 to 50 percent of the *C. melanura* females were engorged. However, during the interval after the pheasant chicks were available, the incidence of blooded *C. melanura* increased to as high as 90 percent. From this it appeared that pheasant chicks were attractive hosts for blood feeding. In addition it may indicate that there is a tendency for the localization of this species around the penned pheasant flock, and may perhaps help explain the reported frequency of virus isolation from collections of *C. melanura* taken at the sites of domestic pheasant flocks.

In laboratory study of the host-feeding of *C. melanura,* experimental engorgement with blood was obtained from chicken egg embryo, juvenile pheasants, chicks, rats, rabbit, white mice, red-winged blackbird and man. In all cases the feeding activity occurred only in subdued light and was a prolonged process characterized by many probings before blood was slowly taken. The easily disturbed feeding was, as described by Chamberlain and coworkers (5), and by Hayes and Doane (11), such a tedious process that many probing attempts were usually necessary before engorgement occurred. This nervous feeding habit probably accounts for the high incidence of mixed blood meal composition reported for this species, and as Hayes and Doane (11) point out, may be an important factor in its role as a virus vector.

SUMMARY

Culiseta melanura is a mosquito which has become of special interest since it is the only arthropod from which EEE virus has been repeatedly isolated. While circumstantial evidence has been reported indicating it may be a vector of the EEE virus either in endemic or epidemic cycles, the distribution of this species in Connecticut has not correlated with the distribution of farms where there has been virus activity. Studies indicate that *C. melanura* will take blood from a wide range of hosts but is not persistent in its feeding activity.

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SOME EFFECTS OF SCREENS IN RETARDING ENTRY OF THE COMMON SALT-MARSH SAND FLY *CULICOIDES FURENS* (POEY) (DIPTERA: HELEIDAE) [1, 2]

JOHN E. PORTER

Culicoides furens (Poey) is widely distributed in the Americas. It is found as far south as Brazil and as far north as Massachusetts. It is prevalent in the West Indies and in the coastal areas of the Atlantic Ocean and the Gulf of Mexico in the United States. It is the most abundant species of *Culicoides* in Florida where it ranks high among the major pests of man.

The blood-sucking habits of these insects attract them to man. Their bite, in many people, produces a lesion more painful and of longer duration than that of some of the pestiferous mosquitoes from the same regions. They do not transmit disease organisms in the United States, but are known to be the intermediate hosts of *Mansonella ozzardi* (Manson) in the British West Indies (Buckley 1934).

The problem created by large numbers of them entering dwellings is one which taxes the resourcefulness of home owners and those people interested in furthering real estate and tourist developments. It is an important item in the budgets of the district mosquito control units which annually spend large sums of money in attempts to protect the populace from these pests. Sand fly control at drive-in theaters is a must to insure maximum attendance. Attendance at public parks is considerably reduced during the periods of peak sand fly abundance.

Prior to 1949 the principal measures directed at larval control consisted of the diking and pumping of salt marshes to prevent tide water overflow and rainwater retention. These measures are only partially successful because little was known then, or even now, of the biology of the species.

When these studies, concerning screens, were initiated in 1950 the outlook was not yet too promising for the use of the newer insecticides as sand fly larvicides, while space spraying or fogging for adult control was prohibitive in most areas because of excessive costs. No evaluation had been made of screening for the exclusion of *Culicoides,* although numerous implications had been drawn. There has been very little scientific information published on the effectiveness of screens or of the chemical treatment of screens to retard insects. It was the author's opinion at this time that an evaluation of insect screening effectiveness could well be made. The periodic peaks of great abundance and the regularity of high populations for several months of the year in the Miami, Florida, area, made the use of *C. furens* the most suitable of the Heleidae for use as a test insect. The following experiments were conducted from 1950–1955, in an attempt to shed light on the effectiveness of those screening materials most commonly in use around dwellings and offices.

MATERIALS AND METHODS. The relative numbers of sand flies trapped in live animal cages was the method used for ascertaining the effects of different types of screening in retarding sand fly entry. In these tests, cages containing rats were provided with 4 "windows" comprising a total of 72 square inches. These windows were screened with one of a series of seven different types of screening.[3]

[1] From the Division of Foreign Quarantine, Public Health Service, U.S. Department of Health, Education, and Welfare, Quarantine Station, Miami Beach, Florida.

[2] A portion of a dissertation presented in partial fulfillment of the degree of Doctor of Philosophy at the University of Illinois, 1955.

[3] The author expresses his gratitude to Mr. R. W. Bacon, Insect Wire Screening Bureau, N.Y.C. for supplying the wire screen materials tested, and to The Chicopee Manufacturing Corporation of Georgia, for the plastic screening used in tests.

The screen materials used were:

Galvanized insect wire screening, 14x14 mesh—0.011 inch diameter wire; Bronze insect wire screening, 14x18 mesh—0.011 inch diameter wire; Aluminum insect wire screening, 14x18 mesh—0.013 inch diameter wire; Bronze insect wire screening, 18x18 mesh—0.011 inch diameter wire; Aluminum insect wire screening, 20x20 mesh—0.013 inch diameter wire; Plastic insect screening, 14x18 mesh—0.015 inch filament diameter; Plastic insect screening, 20x20 mesh—0.015 inch filament diameter.

The word "mesh" is used in the above designation of the various screen materials, and elsewhere in the text, to mean the number of open spaces per linear inch in both warp and filler directions of the screen.

Three replicates of each test were run, with each replicate consisting of a check cage without screen and one cage each in which the "windows" were covered with one of the seven screen materials being evaluated.

Each replicated series of cages was placed in an area of the salt marsh with similar conditions of shading, light, wind currents and attractiveness to sand flies.

The cages used were modified live animal traps designed by Curran and Goulding (Figure 1) (Personal communication, 1951). Each contained one domesticated rat within an inner wire cage. Entering the cages through the screen barrier being

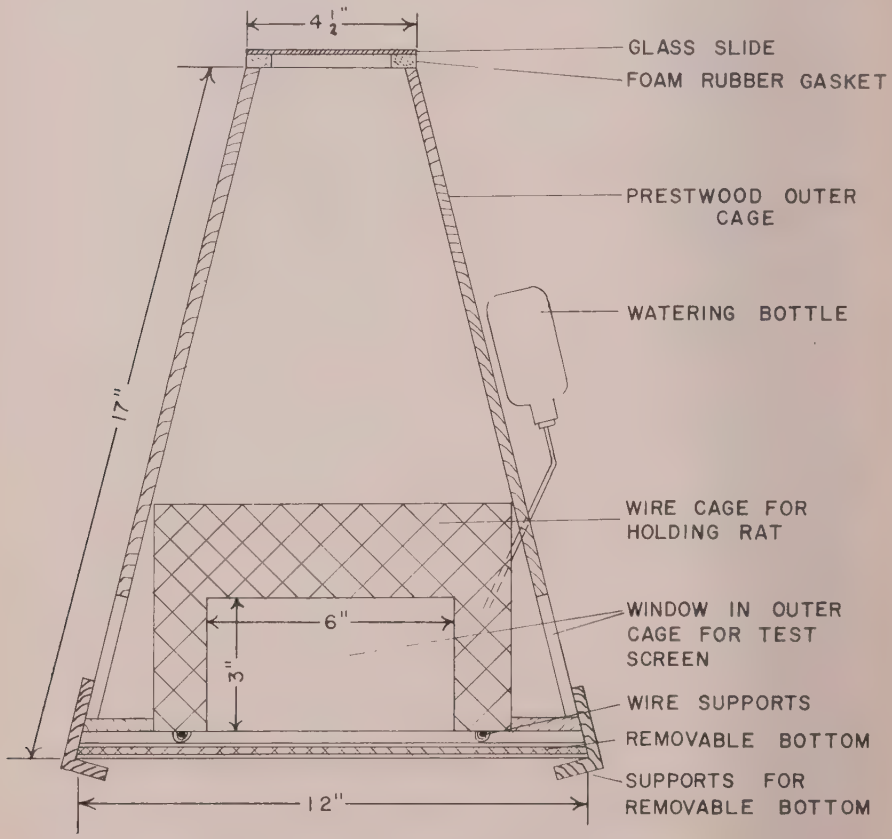

FIG. 1.—Sand fly trap showing construction details.

evaluated, the sand flies would feed upon the rat and seek to leave the trap. The top of the cage was covered by a glass plate coated on the underside with tanglefoot. Flying upward, the insects were caught in this plate. The comparative rate of sand fly entrance retardation for the trap was then determined by the relative numbers of sand flies trapped on these plates.

Daily counts were made of the numbers of sand flies caught on the glass plates. A binocular dissecting microscope was used in making the counts. A very thin layer of tanglefoot must be used in order to allow for proper identification of *C. furens* from some of the similar but less common *Culicoides* of the area, such as *C. inamollae* Fox and Hoffman, *C. canithorax* Hoffm. and *C. melleus* Coquillett.

Frequently extremely large numbers of sand flies occurred on a single glass plate. In these cases only a quadrant of the slide was checked and the total for the slide arrived at by multiplying by four the number found in the quadrant. It was convenient to have a quarter-inch grid paper beneath the slides as a guide in systematically determining the counts in all cases.

The light passage properties of the various screens were simply arrived at by using a darkened wooden box (Figure 2), open on one end and with a narrow slit at the other end. The open end was covered by the screen to be tested while the light receptive gauge of an exposure meter was placed in the slit at the opposite end. A centrally placed 60-watt light bulb was the standard source of illumination. Readings made without screening in the box and with screens in place, gave the data necessary to determine the light passing ability of the screen in question.

Airflow determination for the different screens was obtained by placing a sample of the screen in question over a 3 inch x 3 inch opening at the end of a pyramidal cone. At the large open end of this pyramid a fan was used to create a regular wind flow. An anemometer was placed two inches in front of the screen, so that the screen was between the wind source and the anemometer (Figure 3). Readings made with and without screening in place gave the comparative data necessary to determine the ability of the various screens to retard wind passage.

CALCULATION OF PERCENT ENTRANCE RETARDATION. It was recognized that the different traps would vary in their acceptability to sand flies for entrance and that this variance would, in large measure, be due to location, shading or shelter of the trap from wind. Another factor would be the density of larval breeding in the surrounding area. In the experiments, possible changes in entry acceptance capacity of

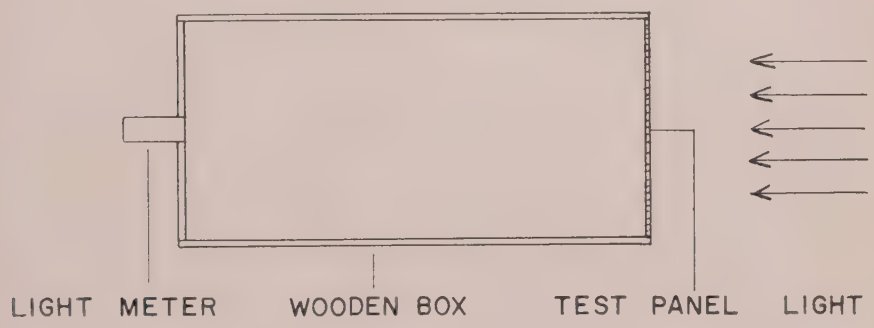

FIG. 2.—Device used to evaluate light penetration through screening.

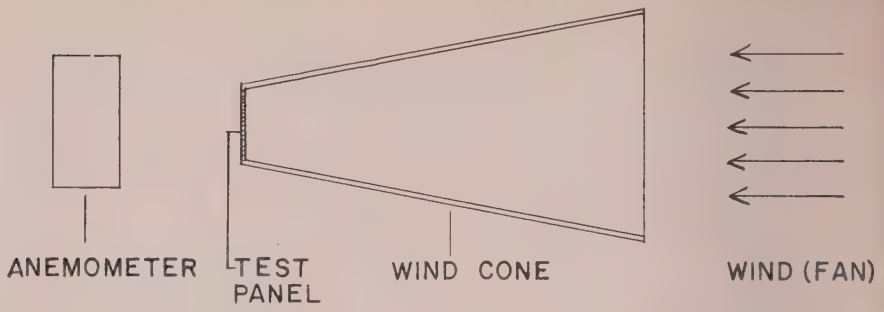

FIG. 3.—Device used to evaluate air flow through screening.

the traps were considered in utilizing the daily sand fly counts from all of the traps during a pre-treatment period when none were screened.

Entrance acceptability of the traps was determined by averaging 12 counts, replicated 3 times (12-day count) on each trap. If the pre-treatment counts on the unscreened cages vary from their counts during the tests, it is assumed that the same forces will be acting on the screened cages. Therefore, the entrance acceptance counts on the screened cages are corrected proportionately upward if more sand flies are found on the checks, or downward if fewer sand flies are observed. This is termed the "corrected normal count" and may be represented by the formula:

$$\text{Corrected normal count} = \frac{\text{—Avg. entrance count on checks during test period}}{\text{—Avg. entrance count on checks during pre-test period}} \times \text{Entrance acceptance counts of screened cage (or screen treatment)}$$

Percent sand fly entrance retardation is evaluated by comparing the corrected normal count with the treatment, thus:

$$\text{Percent entrance retardation} = \frac{\text{corrected normal count—treatment count}}{\text{corrected normal count}} \times 100$$

TEST RESULTS. In general, the results of the tests (Table 1) indicate that screening, even of the best available quality, is not satisfactory in itself for the complete exclusion of *C. furens*. The most satisfactory screening was 20x20 mesh plastic, excluding 96.3 percent of the sand flies; 20x20 mesh aluminum excluded 95.1 percent. The small apertures in these screens, however, also cut down on the airflow and light passage to a point where, in the semitropics, they are no longer satisfactory for use on buildings. These undesirable traits precluded the testing of screens of finer mesh, even though it can be assumed that screening of smaller aperture size probably would be sufficient to completely exclude these pests.

Of the remaining screen types tested, 18x14 plastic and 18x18 bronze were found to be the most practical. The number of sand flies excluded is reasonably close to that of the 20x20 mesh screening and yet the rate of airflow and light passage is materially greater.

However, it is entirely possible and indeed probable that either of these two screens would actually allow for sufficient sand fly entrance in dwellings to cause considerable annoyance and irritation, especially when the sand fly population is extremely high.

The larger numbers of sand flies penetrating 18x14 mesh aluminum, 18x14 bronze and 14x14 galvanized screening, eliminate them from further consideration.

Table 1 shows that in all of the screen

TABLE 1.—Exclusion factors of various mesh untreated screens used for sand fly retardation

Type screening and mesh in inches	Average size of screen openings (sq. inches)	Percent of light penetrating screening	Percent of air penetrating screening	Percent retardation of sand flies
Bronze 18x14	0.002694	65.3	75.4	83.4
Plastic 18x14	0.002288	55.7	70.3	94.6
Aluminum 18x14	0.002485	61.6	75.6	56.1
Galvanized 14x14	0.003648	67.3	80.2	45.5
Bronze 18x18	0.001895	63.8	71.5	90.0
Plastic 20x20	0.001444	50.0	65.2	96.3
Aluminum 20x20	0.001369	49.7	64.2	95.1

types tested there is a close correlation between the number of sand flies excluded and the amount of air and light passing through the screens, with the exception of aluminum 18x14 mesh screening. It can be surmised that the failure of this screening to exclude a larger number of sand flies than 18x14 mesh bronze screening which has larger apertures, is due to color differences of aluminum and bronze. The sand fly's reaction to these colors probably accounts for the greater degree of retardation by bronze over aluminum screening.

The literature reviewed and the results of the experiments herein reported indicate the necessity for fine mesh screening if 100 percent sand fly exclusion is to be obtained. Yet, the evidence seems pronounced that regardless of adoption of a screen with a specific aperture size, some few insects will penetrate. Witness the penetration of recommended screening by small sized *Aedes aegypti* (L.) (Block 1945, 1946), (Stearns and Gillespie, 1945), or the need to specify 16x16 mesh screening as a barrier to *Anopheles gambiae* Giles and *A. funestrus* Giles in West Africa (Davey and Gordon, 1938). The measurements of these mosquitoes are considerably greater (over 0.166 inch) than those of a specimen of *C. furens,* which measures only 0.0018 inch dorso-ventrally and 0.0018 inch from wing base to wing base. Theoretically, a screening of horizontal and vertical proportions slightly less than this would bar entry of all of these sand flies. This, however, would be prohibitive since the mesh would be too fine for practical purposes.

Summary. Seven different meshes of insect screens namely, 14x14 galvanized, 18x14 bronze, 18x14 aluminum, 18x14 plastic, 18x18 bronze, 20x20 plastic and 20x20 aluminum were evaluated as barriers to the entrance of the common salt-marsh sand fly, *Culicoides furens* (Poey). Four screens were found to exclude 90 percent or better of the sand flies, 20x20 aluminum, 20x20 plastic, 18x18 bronze and 18x14 plastic, but two of these, 20x20 aluminum and 20x20 plastic, excluded so much light and airflow that they are not ordinarily satisfactory for use in the tropics or semi-tropics.

The rate of airflow and light passage was determined for the seven screen materials tested. Its relationship is proportionate to the numbers of sand flies excluded.

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CULICOIDES BREEDING IN HUMAN SEWAGE SITES OF DWELLINGS IN TEXAS[1]

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In 1955 a program was initiated to develop techniques for colonizing *Culicoides variipennis sonorensis* Wirth and Jones, a suspected vector of blue tongue of sheep. To facilitate collection of these insects, it became necessary to locate natural areas of heavy, concentrated breeding. Surveys were made of numerous types of environments in the vicinity of Kerrville, Texas. With one exception, the only breeding places that contained large numbers of *sonorensis,* sufficiently concentrated for rapid collection, were on ranches at the outlets of disposal systems for human sewage. Observations on the characteristics of these sites and the species breeding in them are presented in this paper.

GENERAL FEATURES OF BREEDING SITES. In this paper a human sewage site refers to the wet area at the outfall of a drain pipe from which water-borne human sewage or sewage effluent flows out onto the ground. In this type of site, the water disappears into the ground within a short distance of the outlet, owing to the relatively small amounts of water discharged, as these sites are associated with single dwellings for a few people.

Apparently many of these sites on Texas ranches are septic tank overflow areas or, possibly, disposal areas for fluid waste from sources other than the toilet, since raw, whole excreta were rarely noted at the drain openings.

Psychodids were always present in these sites. However, in areas of dense *sonorensis* breeding they were found in small numbers. In several sites that had very large psychodid populations, no specimens of *Culicoides* were found. Several sites of dense psychodid breeding had a much heavier cover of algae than was found on good *sonorensis* sites.

The extent of the sites studied varied considerably, depending on such factors as the terrain, the rate of water flow, the porosity of the soil, and the amount of weed cover present. The area encompassed by each of the three sites of heavy breeding was 30 to 40 feet in length, with the first 10 to 20 feet consisting of a narrow ditch.

INDIVIDUAL SITES. (1) Edwards County, Substation No. 14, Texas Agricultural Experiment Station, human habitation, numerous observations from August 1955 to December 1957. This site apparently resulted from a septic tank overflow, as raw sewage was not evident at the drain opening.

Since there was no cover of weeds, the mud surfaces were exposed to direct sunlight. In 1955 this site consisted of a narrow, shallow ditch that led to areas of exposed mud 20 to 40 feet down a slight slope. These mud areas were surrounded by a very boggy carpet of Bermuda grass that was about 6 feet in width. Two large central areas, each 1 to 2 square yards in size, were completely free of grass and contained loose, brown, fine silt several inches in depth. Densest breeding occurred in these areas and consisted almost entirely of *sonorensis,* although a few specimens of *crepuscularis* were always present. In these two exposed pools the mud was never covered with water deeper than ½ inch. Some mud protruded shallowly above the water in addition to the shallow mud banks. Much of this exposed mud had a moderate covering of algae.

The occurrence of culicoid breeding in this site was briefly described by Wirth and Bottimer (1956).

In a similar area close by, but completely

[1] The following personnel of the Kerrville, Tex., laboratory assisted in the work reported in this paper: H. M. Brundrett, R. R. Kott, M. O. Pickens, and D. E. Hopkins.

and heavily grassed, slight breeding of *sonorensis* occurred in the mud of the boggy turf in 1957. Moderate breeding had occurred prior to 1956 in a few small areas of exposed mud that at that time were not overgrown with grass.

(2) Gillespie County, ranch house, numerous observations from July through December 1957. Fresh human excrement was present at the drain opening at all times. This site ranged from about 30 to 40 feet in length and was free of weeds. The first 10 feet from the outlet consisted of a shallow ditch about 1 foot wide. There was considerable water present, with a pool about 6 inches deep at the drain's exit.

Some larvae were found in the fresh sewage at the drain opening, and they became numerous at the beginning of the black silt banks about 6 feet distant. Densest breeding occurred in these mud banks and in small areas of exposed silty mud in the moderately heavy carpet of Bermuda grass that began at the point where the main channel was diverted into several shallow flows. Later in the year, with an increase in the rate of water flow, heavy breeding became established farther down the gentle drainage slope in a shallow layer of silt that was heavily covered with algae.

As material used for transmission studies was obtained from this site, and therefore a large quantity of flies had to be carefully determined, an opportunity was presented to determine the sex ratio of *sonorensis* and the kind and number of other species present. Of the total of 10,421 specimens determined from randomly selected groups of flies that emerged over a 2-week period, 10,354, or 99.4 percent, were *v. sonorensis*, 61 percent of which were females. In addition, 65 specimens of *crepuscularis* and 2 of *haematopotus* were present.

(3) Kerr County, ranch hunting cabin, numerous observations from July 1956 through June 1957. Fresh feces were not evident at this site. It was ascertained that the fluid waste passed through a septic tank.

This site was about 30 feet long. The first 15 feet consisted of a moderately deep ditch with a narrow muddy area at the bottom. The second 15 feet, where the ditch broadened out, was a flat, oval area well covered with Bermuda grass but interspersed with small muddy spots caused by the trampling of livestock. Cow manure was usually present in small amounts. This site was dry during the drought of 1955. In 1956 and the spring of 1957 the water flow was sufficiently strong for livestock to drink. A weed growth was not present in 1956. However, a heavy cover of weeds developed by June of 1957 and, because of the subsequent shading and reduction in water flow, heavy breeding no longer occurred.

This site was unusual in that it was not characterized by areas of loose, silty mud free of grass. Breeding was heavy in the intermittently muddy sod of the lower flat area and in the muck of the narrow upper ditch. Moderate to heavy breeding was found in 1956, and collections were made in May, July, and September. In 1957 moderate breeding was discovered on February 6, and heavy breeding was first noted on March 19. On this date thousands of larvae could be collected by merely scooping them from the clear water in the lower flat area where it was as deep as 1 inch. At this time larvae were also commonly noted in the shallow, clear, running water, maintaining a static position by swimming against the currents, above the mixture of leaves and mud contained in the narrow upper ditch.

This unusual activity of the larvae in the clear water was apparently the reaction of a high population to an inadequate food supply. This conclusion is strengthened by the fact that the size of collected full-grown larvae diminished during this 2- to 3-week period. Those collected in March were retained by a 50-mesh screen, but in April larvae ready to pupate passed through the 50-mesh screen but were retained by a 60-mesh screen. This lessening in size of collected full-grown larvae was believed to be a response to overcrowding, with the resultant scarcity of nutrients, as there was probably not sufficient time for a seasonal effect to take place.

On the morning of April 24, 1957, a 4.4-inch rain fell very rapidly. As this site is close to the bottom of a shallow draw, it was covered by the swiftly flowing water to a depth of several feet and most of the larvae were swept away. However, about 2 weeks later a few full-grown larvae were found, indicating that some had survived the torrent, probably in the mud held in place by the sod.

Two collections of pupae were made to determine the number of each species present. In addition, a mud sample for laboratory emergence (March 4, b) was taken. Results are given below.

		variipennis			*crepuscularis*		*haematopotus*
Date 1957	Total	Total	♀♀	♂♂	♀♀	♂♂	♀♀
February 6	100	96	—	—	4	—	—
March 4							
Sample a	91	91	53	38	—	—	—
Sample b	96	94	50	44	—	1	1

The occurrence of *sonorensis* breeding at this site was mentioned by Wirth and Jones (1957).

(4) Gillespie County, ranch house, July 1957. Fresh excreta were evident at the mouth of the drain. Moderately heavy breeding of *sonorensis* was found at the mud margins close to the drain opening. A cover of Bermuda grass was not present. Breeding was not found farther down the slope where there was a heavy growth of weeds. There was little silt at this site, and one moderately large collection of silt reduced breeding to almost nothing.

ABUNDANCE OF BREEDING SITES AND SEASONAL OCCURRENCE. Heavy-breeding areas of *sonorensis,* either small sites of dense breeding or large sites of moderately concentrated breeding, were not common in the Kerrville area during the last few seasons. A survey of ranches in midsummer of 1957 covered 69 sites that appeared to be suitable for breeding of this species. Of these, 26 were human-sewage effluent sites. One heavy and one moderately heavy breeding site were discovered, both human-sewage sites. Other sites had slight or no breeding of *sonorensis* at the time of sampling.

The time of year at which populations of *sonorensis* were heaviest was not ascertained, because large quantities of material were constantly being removed from the breeding sites for laboratory experimentation, but it was evident that population levels were lowest during the winter months.

ECONOMIC CONSIDERATIONS. Heavy, concentrated breeding of *variipennis sonorensis* was not commonly found on ranches, and most of the flies found on a ranch could be controlled easily by the treatment of one small site. Control at the ranch house could be very important, as this area is frequently the pivotal point of ranching operations. On many ranches the animals pass through corrals near the house at least once a year in normal handling, and culicoid breeding in this area could well be a center of animal disease transmission.

An abundance of these flies breeding in human sewage close to habitations might also constitute a human health hazard. Although culicoid flies have thus far not been incriminated in the transmission of human disease in North America, it remains a possibility. Whitehead (1934) has shown that under outbreak conditions *variipennis* prefers livestock to man. According to Wirth and Jones (1957), P. J. Spangler found that in Missouri man is not a preferred host of *variipennis australis* Wirth and Jones, the subspecies associated with saline environments.

Laboratory tests at Kerrville indicate that *sonorensis* is not host-specific. It readily engorges with blood from cattle, sheep, rabbits, and mice. In one test with a few flies, two females readily fed on the foot of a chicken. In a small series of tests, in which a Jersey bull calf, a ewe lamb, and the author as observer were confined in a cubical cloth cage with released flies, they

showed preference for cattle, sheep, and man in descending sequence. The author received 8 bites in comparison with 42 observed engorgements on the calf. Under certain conditions this species might constitute a health hazard to man, especially in the absence of a preferred host.

Summary. Four good breeding sites of *Culicoides variipennis sonorensis* Wirth and Jones were found near Kerrville, Texas, three with a heavy and one with a moderate population. These sites were human-sewage disposal areas at dwellings, and were the result of allowing water-borne sewage or sewage effluent to flow out onto the ground. Two of these four sites were apparently created by septic tank effluent. The other two resulted from the discharge of raw sewage, as evidenced by the presence of whole excreta by the drain opening.

One factor associated with heavy breeding was the presence of a heavy carpet of Bermuda grass instead of weeds, with consequent exposure of the mud surfaces to direct sunlight. Larvae were not found in areas that were thickly covered with weeds. Breeding was heaviest in nonvegetated areas of exposed, silty mud, such as shallow banks close to the drain opening, and open pools of mud surrounded by the turf that carpeted the drainage slope.

At least small numbers of psychodids were always present in association with *sonorensis*. A dense population of one apparently precluded a large population of the other.

Heavy-breeding sites of *sonorensis* were scarce in the vicinity of Kerrville. A survey of ranches in midsumer of 1957 covered 69 sites that appeared to be suitable for breeding of this species. Of these, 26 were human-sewage effluent sites. One heavy and one moderately heavy breeding site were discovered, both human-sewage sites. Other sites had slight or no breeding of *sonorensis*. Breeding occurred throughout the year, but population levels were lowest in the winter. Apparently *crepuscularis* and *haematopotus* usually occur along with large populations of *sonorensis* in these sites, but in such small numbers that their presence is frequently concealed by *sonorensis*.

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LIGHT TRAP INDICES OF MOSQUITO ABUNDANCE: A COMPARISON OF OPERATION FOR FOUR AND SEVEN NIGHTS A WEEK

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Many time-saving techniques have been tested and applied to the task of processing mosquitoes captured in light traps and other collecting devices. A large part of the work, however, is that of separating mosquitoes from the mass of other insects collected. This problem assumes major proportions when light traps are operated each night throughout the season. Several investigators have concluded that useful data indicating population trends of adult mosquitoes can be obtained from the operations of light traps on a limited schedule (Bradley and Travis, 1942). J. A. Mulrennen and D. M. Rees (personal communications) found that the operation of light traps for two nights a week was sufficient to follow population trends of mosquitoes in Florida and Utah, respectively. For the present study a four-night schedule was selected because it allowed the longest time for operating a light trap consistent with a five-day work week. On the fifth day, Friday, the collections were processed and results tabulated.

MATERIALS AND METHODS. Light trap collection data from mosquito control districts in two regions of the Central Valley of California were evaluated. These districts were selected because their records were kept on a daily basis. American model light traps (Mulhern, 1953), with 25 watt, white, inside-frosted bulbs, were used in all instances. Data for 1954 were obtained from seven districts; three from the northern and four from the southern regions of the Central Valley. Data from five of the seven original districts were utilized in 1955 as well as records from two traps operated by the Bureau of Vector Control in the southern region of the Valley. All data were analyzed first on a seven, then on a four trap-night per week basis. The four trap-night data utilized Monday through Thursday night collections. The analysis was limited to the numbers of female *Culex tarsalis* Coquillett collected. This species is normally the most common mosquito taken in light traps throughout the Valley during the summer months. Records for the species are tabulated separately because of its importance in transmission of arthropod-borne encephalitis. The analysis was limited to females since some districts do not record males either because of difficulty of identification or the low numbers collected. All figures are expressed as average number of female *C. tarsalis* per trap night.

The unit of measurement was the trap night. This is defined as one trap operating for one night, from 6 p.m. to 6 a.m. Light trap figures for the seven-night operation were obtained by dividing the total weekly catch for all traps operated in each district by seven times the number of traps operated; for the four-night operation the trap night figures were obtained by dividing the total catch for all traps by four (Monday through Thursday nights) times the number of traps operated.

For the comparison of indices based on trapping for four or seven nights, a standard error of the difference was calculated which took into consideration the high degree of correlation between the two sets of measurements.

RESULTS. There were 14 sets of data which were compared with respect to four or seven nights' trapping. Of these, eight sets are shown in the accompanying table and figure. It will be noticed from the figure that there was relatively little difference in the indices based on four and seven nights a week.

Figure 1 also shows four comparisons

Figure 1

NUMBER OF FEMALE CULEX TARSALIS PER TRAP-NIGHT
COLLECTED FROM SELECTED LIGHT TRAP LOCATIONS
IN THE CENTRAL VALLEY, CALIFORNIA, JUNE-SEPTEMBER, 1954-1955

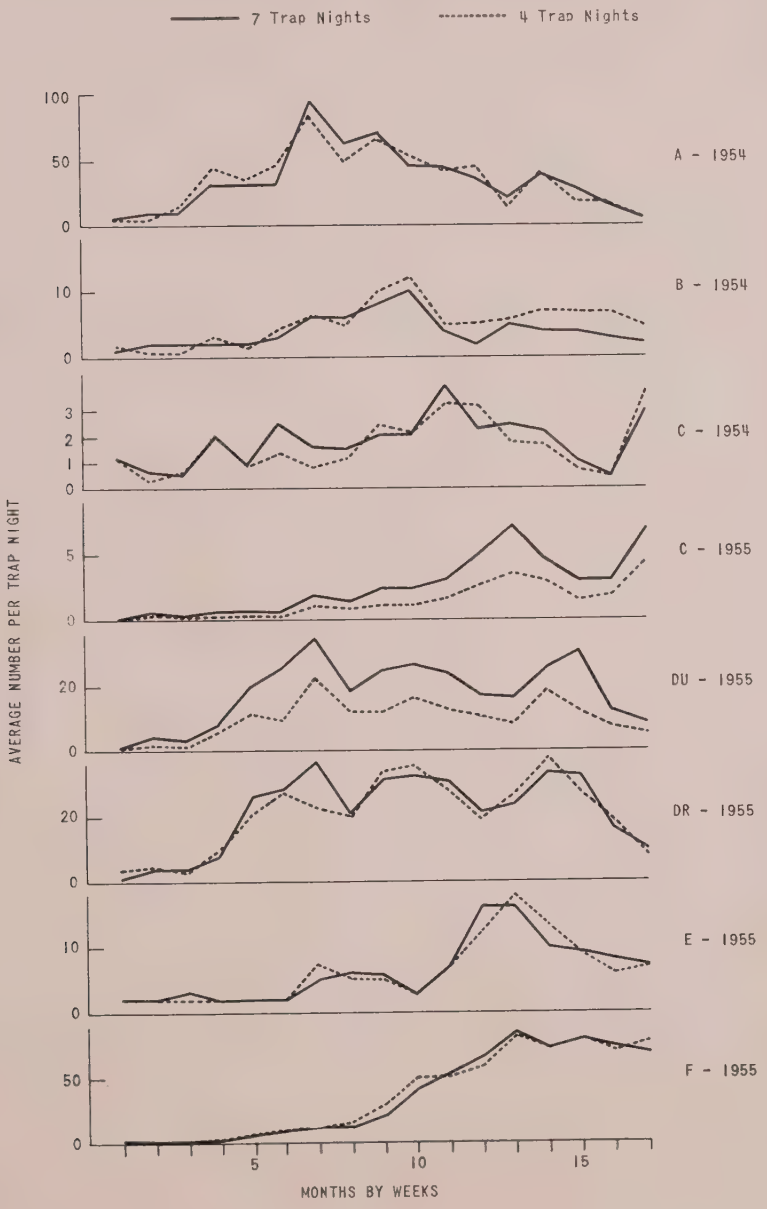

TABLE 1

NUMBER OF FEMALE CULEX TARSALIS PER TRAP NIGHT
COLLECTED FROM SELECTED LIGHT TRAP LOCATIONS
IN THE CENTRAL VALLEY, CALIFORNIA, JUNE-SEPTEMBER, 1954-1955

DATE		LOCATION															
		A		B		C-54		C-55		DU		DR		E		F	
		NUMBER OF TRAPS															
		1		8		5		5		6		6		1		1	
		NUMBER OF NIGHTS															
		7	4	7	4	7	4	7	4	7	4	7	4	7	4	7	4
June	5	1.8	2.0	0.9	1.7	1.3	1.2			0.7	0.4	0.9		1.5	2.3	1.5	1.7
	12	6.5	3.7	1.4	1.3	0.7	0.3	0.6	0.4	3.7	2.1	4.4	4.6	2.0	2.5	0.3	0.2
	19	9.3	13.2	1.3	1.3	0.5	0.6	0.3	0.2	2.8	1.0	4.4	3.4	2.7	2.5	1.3	1.3
	26	31.3	43.0	2.2	2.6	2.0	2.0	0.7	0.3	8.0	5.7	7.9	9.2	2.3	2.2	1.3	1.0
July	3	32.0	34.2	2.2	2.1	0.8	0.8	0.7	0.4	19.2	11.0	25.9	21.2	1.6	2.0	6.3	7.5
	10	32.5	44.0	3.9	4.3	2.5	1.3	0.6	0.3	24.9	9.3	28.0	27.1	1.5	1.7	8.1	10.0
	17	93.5	84.0	5.7	6.2	1.6	0.8	1.8	1.1	34.3	22.2	37.5	32.6	5.1	7.2	6.1	9.8
	24	62.1	45.7	6.4	5.2	1.5	1.1	1.5	0.9	18.5	11.8	20.7	20.5	6.4	5.2	12.0	14.0
	31	70.2	65.0	9.2	10.0	2.1	2.3	2.5	1.2	24.2	11.8	31.2	33.6	6.0	4.8	21.0	28.2
August	7	47.0	52.0	9.7	13.6	2.0	2.1	2.4	1.2	26.2	16.1	32.4	35.0	3.1	3.2	41.0	49.7
	14	45.7	43.0	4.6	5.0	3.8	3.2	3.1	1.7	23.6	12.4	29.9	28.3	7.1	7.1	53.7	52.7
	21	36.5	44.0	4.0	5.1	2.3	3.1	5.0	2.6	17.4	10.0	21.3	19.3	16.0	11.5	64.0	57.5
	28	21.1	14.0	3.9	5.0	2.4	1.8	7.2	3.4	16.7	8.2	22.9	26.1	16.0	17.5	82.7	81.0
September	4	37.8	39.0	5.0	5.6	2.2	1.6	4.7	2.9	25.5	17.6	32.9	38.3	10.0	12.8	71.9	72.7
	11	28.2	18.5	5.2	6.8	0.9	0.7	3.0	1.5	29.8	11.7	32.1	27.5	9.0	9.0	78.0	78.0
	18	15.7	17.0	5.2	6.7	0.5	0.5	3.0	1.8	11.6	6.8	16.0	18.1	7.6	6.0	72.4	70.2
	25	3.8	4.0	3.0	4.6	2.8	3.6	6.7	4.4	7.7	4.9	9.0	8.8	6.9	7.0	67.1	76.5
Total		575.0	566.3	73.8	87.1	29.9	27.0	43.8	24.3	294.8	163.0	356.5	353.6	104.8	104.5	588.7	612.0
Mean		33.8	33.3	4.3	5.1	1.8	1.6	2.7	1.5	17.3	9.6	22.3	22.1	6.2	6.1	34.6	36.0
Correlation Coefficient		0.952		0.952		0.854		.973		.929		.962		.936		.993	
Standard Error of Difference		1.84		0.197		0.118		0.251		1.23		0.781		0.396		1.443	

Source: State of California, Department of Public Health, Bureau of Vector Control Records.

which indicate that the four-night index is satisfactory under a variety of conditions. Areas A and B represent districts in the northern and southern regions, respectively, of the Central Valley in 1954. Seasonal trends in 1954 and 1955 are shown by District C located in the Southern Central (San Joaquin) Valley. Trends in urban (DU) and rural (DR) areas are shown from a district in the northern Central (Sacramento) Valley in 1955. Examples E and F show trends of *C. tarsalis* populations in areas of the San Joaquin Valley in 1955 where mosquito control measures were or were not practiced, respectively. Comparable results were obtained from the other six sets of data which are not included for the sake of brevity.

Table 1 shows the the average number of mosquitoes per trap night for eight trap locations for seventeen weeks, on a four- and a seven-night basis. The means, the correlation coefficients, and standard error of the difference of the means are shown in the lower part of the table. For many individual weeks the number of mosquitoes per trap night for a given location are very similar for both four and seven nights. The mean number of mosquitoes per trap night for the entire season differed significantly in three of the eight sets of data (B, C—55, DU). The difference in a fourth location (C-54) approaches significance at the 95 percent level. Thus it can not be concluded that there is no difference between indices based on collecting for four or seven nights. The high degree of correlation between these schedules of operation, however, shows that for most purposes the four-night schedule is satisfactory.

Discussion. These results closely parallel those of Bradley and Travis (1942) who found that "the mean daily catches, obtained from trap operations made every second and third day, approximate closely to the mean catch from traps operated every day." These workers found that the magnitude of the standard error increased as the interval between sampling increased; the standard error for an index based on catches made every third day was approximately twice the standard error of the index based on daily catches.

In view of the many factors that may affect light-trap catches, such as temperature, humidity, wind direction and velocity and the relative effectiveness of mosquito control in surrounding areas, the authors believe that these results justify the use of *C. tarsalis* indices based on operation of traps for four nights a week in the Central Valley of California.

Summary. Operation of light traps on either a four- or seven-trap night a week schedule was shown to give acceptable seasonal population trends of adult *Culex tarsalis* in the Central Valley of California. The two schedules of operation gave similar results under a variety of conditions and in different locations during a two-year period. Light traps used four consecutive nights, Monday through Thursday, rather than seven nights may insure a better schedule of operations within mosquito control agencies.

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OBSERVATIONS ON ALASKAN SNIPE-FLY PESTS (*Symphoromyia:* Rhagionidae)

W. C. FROHNE [1]

INTRODUCTION

Two common Alaskan species of *Symphoromyia,* viz. *atripes* Bigot and *kincaidi* Aldrich, are the biters *par excellence* of the only North American biting genus of rhagionids, or snipe flies. There exist local pest problems attributable to *atripes* in subarctic Central Alaska and to *atripes* plus *kincaidi* in temperate Southeastern Alaska. Travis (1949), and Sailer (1951) reported *S. atripes,* and Frohne & Williams (1951) added *kincaidi* and sketched distribution, habitats, and the biting habits of both species. This notorious Alaskan pair of blood-suckers belongs to an otherwise virtually innocuous and therefore neglected genus of not less than 22 species in the United States and Canada. The Alaskan species, at least, being the bad actors, require investigation: the secretive immatures remain entirely conjectural; the conspicuous females will not be ignored; the mysterious males invite attention. For heuristic reasons, then, to interest entomologists visiting Alaska, my son and I offer our summer snipe-fly notes, a by-product of a major interest in mosquitoes.

Data supporting this paper were secured more from opportunity than by plan. Adults were collected intermittently in an extensive habitat of both species, June 19–August 12, 1958, covering most of the local biting season. Landing rates, biting habits, and habitat characteristics were determined as time allowed. The findings relate chiefly to four matters of long-standing uncertainty:

(1) The taxonomic status of a golden-haired *Symphoromyia* collected over forty years ago in the identical area of this study, Sheep Creek, Thane, near Juneau;

(2) Correct species association of males and females;

(3) Habits of females, especially comparative landing rates, influence of weather on activity, and biting habits;

(4) The habitat, particularly its characteristic vegetation.

GOLDEN-HAIRED *Symphoromyia.* When Aldrich (1915) described *S. kincaidi* he studied several Alaskan specimens acquired from Prof. Melander who had taken a very odd one at Sheep Creek where the present observations were made. Dr. Aldrich tentatively assigned this exceptional fly to his new species with the reservation that it differed by having "entirely yellow pile all over" and that it "may be a different species." Leonard (1930) did not comment. Frohne & Williams (1951) confirmed occurrence of typical *kincaidi* in Alaska on the basis of four biting females from Salmon Creek, which is about fifteen miles northwest of Sheep Creek, stating their specimens have the black hair on front and thorax normal to *kincaidi.* In such cases of uncertainty the best evidence is a series of specimens from the original collection site of the specimen in question. Accordingly my son Dick and I collected 140 *kincaidi* females along Sheep Creek in nine lots over a period of eight weeks. (Two hundred twenty-six *atripes* were taken and pinned at the same time.) We also hand-collected from ourselves other samples of both species and swept by net non-biting rhagionids at five other creeks of the Juneau region. Unfortunately, however, we failed to discover a single *kincaidi* with completely yellow pile. So the interpretation of Aldrich's questionable specimen is still uncertain. Is it a faded changeling, a rare variant of *kincaidi,* or a new species?

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Color characters only are known to separate females of the two species, and the easiest one to use under a lens is pigmentation of halteres, yellow in *kincaidi,* but tan, dark-brown or black in *atripes.* For tentative field identification grey females are *atripes;* brown ones, *kincaidi.* In selecting specimens to pin, which we usually did while counting landing rates, we noticed size and color deviations. Size varies in both species, and we took, e.g., two dwarf females of *atripes* decidedly less than half usual size. Variations in color and pollinosity of the body and infuscation and color, especially in the anal angle of the wings, conspire to make an occasional *kincaidi* almost indistinguishable in the field from *atripes.* On the other hand, even under the microscope I found several flies pre-sorted in the field to *kincaidi* which possess brown halteres. In these questionable specimens the overall color is brownish rather than grey, and the anal angle of the wing is orange. Such specimens will key to *atripes* but should be assigned to *kincaidi.* Abnormalities in wing venation are commonplace in both species. The "open anal cell," a key character of the genus, is closed in about a third of these specimens, though rarely on both wings and there is never much "stalk" distal to such closed anal cells. Color of the pile in *Symphoromyia* is an indefinite, quantitative character tricky to observe properly. Definitely, however, the pile on front and thorax is dark, usually nearly jet-black, in all *kincaidi* we collected at Sheep Creek in 1958.

Association of Males and Females

The proper association by name of the sexes has long been problematical for most North American species of *Symphoromyia.* Aldrich (1915) attempted designation of both sexes for eleven of the twenty-two species. Leonard (1930) completed an additional pair by describing the male of *S. plumbea* Ald. Unfortunately, the chief criterion for applying the same name to male and female forms is but little more than their occurrence in the same area at the same time—collection together. This rule of thumb provides an uncertain basis for *Symphoromyia* because of pronounced sexual dimorphism, intraspecific variation, paucity of diagnostic characters, scarcity of males in collections, and absence of pairs taken *in coitu.* It is usual, unfortunately, in western *Symphoromyia* habitats for more than one species of female to occur on the wing at the same time.

However, we have virtually confirmed Aldrich's association of the sexes of *S. kincaidi.* My son Dick and I swept thirty jet-black non-vittate male *Symphoromyia* at Sheep Creek in 1952. They fitted Dr. Aldrich's description of Washington and British Columbia males he had designated *kincaidi.* Female *kincaidi,* however, were much less common there at the time than *atripes,* and we doubted our new evidence fully confirmed Dr. Aldrich's proper association of male and female *kincaidi.* In 1955, nevertheless, we took nine larger, grey, vittate males resembling female *atripes.* This evidence obtained in the Upper Cook Inlet seems adequate and conclusive proof that the jet male belongs to *kincaidi,* the grey one to *atripes.* Now, Aldrich had based his description of the *atripes* male quite lamely on a single, incomplete specimen collected by Osten Sacken in California in 1876. The Alaskan specimens do not agree well with his designated type, but they seem to have a more legitimate claim to conspecificity with the female everyone calls *atripes* than has Aldrich's male type. For an unknown reason *atripes* males are extremely rare in collections. We have collected most of them (ten) ourselves in Alaska by sweeping. Alexander Hubert, while searching American museums for male *Symphoromyias,* uncovered another specimen matching the Osten Sacken-Aldrich type male *atripes.* In Frohne (1957) these different males are compared. Quite possibly the agreement in swollen first antennal joints of Alaskan males with the Osten Sacken-Aldrich male should outweigh color differences so that both the grey (Alaskan) and darker (Stateside) forms with similarly very swollen first

antennal joints can be considered true *atripes* males. While the *kincaidi* male seems confirmed beyond reasonable doubt there are, nevertheless, two reasons for further collection of *kincaidi* males in Alaska. In the Upper Cook Inlet two jet-black *kincaidi* males were swept along with nine grey, vittate *atripes* males. This is an extraordinary find because no female *kincaidi* whatsoever has yet been collected north of Southeastern Alaska. These two northern *kincaidi* are identical with male *kincaidi,* except one, from Sheep Creek, Southeastern Alaska. The exceptional Sheep Creek male constitutes the other reason for further collecting. This puzzling *Symphoromyia* was swept the same afternoon in 1958 with three normal *kincaidi* males. It resembles *atripes* in some respects, *kincaidi* in others, being brown rather than jet-black, non-vittate, and smaller than most *atripes* males. It is probably *kincaidi* since the first antennal segment is relatively less swollen and carries longer hair than *atripes,* but its wings are completely clear like no other *kincaidi* male of 33 we have taken there. This puzzling specimen quite spoils my assertion (Frohne, 1957) that "*kincaidi* males may invariably be distinguished with the unaided eye from *atripes.*"

Habits of Females

Statements in the literature on snipe-fly biting, pest importance, influence of weather and light on activity, etc. are few and subjective. The blood-sucking habit of *atripes* was firmly established by Aldrich (1915), who was also rightly "under the impression that *kincaidi* is an almost equally bad biter." Frohne & Williams (1951) reported *kincaidi* biting severely in Alaska. In the main, knowledge even of the biting habit has not advanced appreciatively since 1877 when Count Osten Sacken reported an unknown *Symphoromyia* female "stings quite painfully and draws blood like a *Tabanus.*"

The present field notes on biting at Sheep Creek in 1958 relate to fourteen different dates and doubtless give a representative, nearly complete sketch of the seasonal history of the flight period there. The landing rates were determined by averaging counts of flies alighting on the front of the trousers as described by Blanton *et al.* (1950) for mosquitoes. They provide a rough means of comparison with pest problems of mosquitoes and other biting flies.

Snipe flies have been taken biting in the Juneau area from mid-June to mid-September, but the period when they are numerous and active enough to cause a nuisance is much shorter. In 1958 it included all calm, sunny days from June 24 to August 20. These 58 days actually included only about 38 days of bad biting, i.e., with landing rates of five or higher. It is evident that, as for horse flies, people fuss over several snipe flies more than for a score of mosquitoes. The following observations were, to be sure, made in an area of high densities. Yet rates seldom reach five in the Evergreen Bowl, Juneau city park on Gold Creek where public demand requires adulticiding every summer. Hikers to upper Gold and Granite creeks and also to Eagle River encounter higher densities. Salmon Creek, home of Ralph Williams, a keen observer of snipe flies for more than a decade, has equally high densities. Other creeks near Juneau, e.g., Lemon Creek, may be entirely free of *Symphoromyia* although this more likely reflects low densities and inadequate search. We lived on another of these "negative" streams, Eagle Creek, Douglas Island, in 1958 and actually collected a few snipe flies there on three occasions in July.

The first adult *Symphoromyia* of 1958 seen at Sheep Creek were three *kincaidi* on June 19. A half-day search did not show any present June 14. The afternoon of June 24 was sunny, not at all windy, and temperatures on the upper basin level reached 71° F. Both species were present, the combined landing rate varied 2–12, and the sample collection contained 55 *kincaidi* with 38 *atripes.* June 26 was cooler (60° F.) and cloudy, though calm. No landing rate counts were made but it was

noted that a range similar to June 24 characterized the biting activity. However, there was a marked difference in species composition, for few *atripes* were seen. June 30 was a warm, partly sunny day. Landing rates and a sample were taken when a marked increase in *atripes* was noted. The landing rate was 10–20, and the sample was composed of 39 *atripes* and 34 *kincaidi*. July 5 was hot (71° F.), sunny and calm, and snipe flies were very annoying. A cloud of about fifty followed my wife at head height and settled on her whenever she stooped to botanize, the following swarm of the insects much suggesting the behavior of certain *Aedes*. Maximum landing rates were 30, the lowest 10. The following day, July 6, all conditions were similar, and the sample collection shows a pronounced *atripes* preponderance of 60 to 30 *kincaidi*. Several males were swept, and this date was probably near the peak for density of both species, both sexes. July 7 was cool (55°–60° F.) and landing rates were off to about half the previous day's. Similarly, July 9 was cool (55° F.) and flies were still fewer. The sample collection of that day reversed the species proportion, for 22 *kincaidi* to only 13 *atripes* were taken. July 13 was mostly cloudy, but intermittent sunshine raised the midafternoon temperature to 63° F. The landing rates became high late in the afternoon, reaching 25–30. It was especially interesting that *atripes,* evidently more of a fair-weather fly than *kincaidi,* again predominated, 50 in the sample to only 11 *kincaidi*. July 15 was mostly sunny and the temperature climbed to 70° F. Late afternoon landing rates ranged 25–30. None was made earlier but we had the impression of as few as 5 flies. The sample collection contained 52 *atripes,* 11 *kincaidi*. (Cold, windy, rainy weather intervened until July 28.) July 29 cleared, but the upper basin was exceedingly wet. Landing rates were 2–5 in the canyon and 5–10 on the plateau. The *kincaidi* were decidedly scarce and the sample, 23 *atripes* and 5 *kincaidi,* reflected bias from our effort to include the scarcer species. Actually the ratio was probably over 90 percent *atripes*. (Another week of stormy weather interrupted observations.) August 1, the first day of intermittent sunshine, a careful search of the upper basin yielded only a single *atripes* female. It was not possible to return to Sheep Creek in 1958, but we worked at Granite and Gold creeks about two and a half miles to the northwest which have similar upper basins. At Granite Creek landing rates of August 9 and 11, partly sunny days, were 5, and we collected 28 *atripes* to 1 *kincaidi,* and 25 *atripes* to 1 *kincaidi,* respectively. Thus the flies were troublesome on calm, warm days from late June to mid-August, and there was a greater tendency in *atripes* to reduce activity in cool, windy, wet periods, though, in fact, neither species was active in very bad weather.

S. atripes and *kincaidi* bite similarly. However, it appears that *kincaidi* begins biting earlier in June and continues to bite relatively more than *atripes* in rainy, cold, or windy weather. The following notes apply to both species.

Adults are strictly diurnal. Bright sunshine appears to provide optimal light for biting, and activity promptly decreases when clouds obscure the sun. Attack seems to occur in greatest force and most avidly under the open sky. Certainly fewer flies attacked us at Sheep Creek in the coniferous forest, a result, probably, also of increased distance from breeding areas. The first flies are noticed as early as 7 a.m. (AST) on a fair day. Their numbers generally increase gradually, which is to say biting activity increases during the morning. There is a peak about noon on cloudless days, which may be delayed to mid-afternoon on cloudy ones. Lulls with no apparent meteorological cause occurred as in the activities of mosquitoes and other flies. In the study area no sooner does the sun pass below the mountain rim so that most of the basin is shaded all at once than the flies abruptly disappear for the day. In June and July this happens between 7 and 8 p.m. It took place, for example, on June 30, 1958

precisely at 7:45. Within five minutes the last straggler had gone. Not another *Symphoromyia* was seen that day though we worked for an hour longer in the bright light reflected from the blue sky and then hiked another hour on the trail.

Both species are purposeful biters. They alight without preliminary buzzing, that annoying habit of blackflies, and settle chiefly on feet, legs, lower trunk, or arms, less frequently on neck or head. Again differing from blackflies, they are not attracted to eyes and ears, nor drawn like mosquitoes by the breath and warm skin. Selection of host and point of attack is certainly partly visual, for dark-colored clothing, especially olive-drab, is preferred to light-colored and even to the bare skin. Sometimes cloth of woolly (rough) texture was preferred over smoother. However, dark-brown, slightly shiny, smooth leather boots (being worn) were by far the most favored of all. Preference for the posterior half of the human body was still evident when we lay prone or sat under a cliff. About as many flies would land on boots whether one stood or brought the head between the knees and put the arms around the ankles. They occasionally landed on discarded clothing and musette bags but less often, I believe, than on the same articles in use. Even a sweaty white nylon shirt hanging on a bush though of least favored color and texture was not altogether exempted. *Symphoromyias* were not observed to land near a host on ground, rocks or vegetation except once when one crawled on a stone into the spray of the creek. It was collected and dissected, showing no evidence of having fed and no egg development. In fact, except coming to bite or in flight after engorging they were simply not seen in Nature at all. This is not a claim without basis for many hours were spent carefully examining flowers, etc., while miscellaneous Diptera were collected. Visual stimuli are not so important in finding the host as for tabanids which often mobbed our red-brown station wagon, behavior not seen for the more abundant snipe flies.

A reputation for bad biting mentioned by Aldrich and other authors interested us and we made some effort to account for it. Unmolested snipe flies bite repeatedly without taking flight. An *atripes* was allowed to bite my knuckle at the base of the index finger. When about half-engorged it withdrew the stylet, moved about a quarter of an inch and bit again. This was repeated five times until the fly was fully engorged when it flew away. From two to seven minutes per bite or a total of seventeen minutes for the series of six bites elapsed. Four of the bites oozed a little blood and three were momentarily quite painful. In fact I have rarely experienced as sharp pain to provide a full blood meal for a mosquito. The bites were circled in ink and two were rubbed and lightly scratched (although they did not itch), but there was no immediate or delayed reaction, viz. color or swelling, and two hours later nothing but the ink circlets marked the bites.

HABITATS

I have described two Central Alaskan snipe-fly habitats in a previous paper (Frohne, 1957). They are limited foci of five to ten acres each, located at about 3200 feet, well above timber line in the headwaters of Fishhook Creek, Upper Cook Inlet. The terrain is a helter-skelter hotchpotch of wet rocks lushly overgrown along the numerous dashing watercourses by alder, scrubby willows and very tall weeds. On the less rugged lateral slopes and ridges, however, the coarse growth thins out and gradually changes to delicate carpets of alpine herbs and mosses. My wife has furnished a list of characteristic plants. The collection of both species of males in the Fishhook Creek foci and the emergence of two female *atripes* in Dove traps vouch for breeding microhabitat.

The Southeastern Alaskan snipe-fly collecting area at Sheep Creek is on a grander scale, covering nearly all the watershed or not quite a square mile. Highest densities of the females, males, and by inference the larval breeding sources are all found

on a plateau 750 feet above sea level. The females, presumably, disperse a mile or so from the plateau, for they occur at usually progressively lower densities downstream. The upward limits of adult and larval habitats remain conjectural. Perhaps the best surmise as to larval habitat is the inaccessible, wet, alpine cliffs and grassy rockledges between 800 and 1600 feet elevation which enclose the plateau-like palisades, but the only evidence to support this conjecture is the negative one of unsuccessful search for larvae on the plateau. I shall describe our collecting area of predilection one mile along Sheep Creek below Portal Camp on the plateau to orient visiting entomologists seeking male *Symphoromyias*. The local name for the plateau is "upper basin."

Sheep Creek courses 3½ miles from primary sources below Middle Peak in three large, nameless cirques flowing along the so-called upper basin, a U-valley left by the current retreat of the local glacier. Where the glacier presented a sheer face to the sea about 5,000 years ago at elevation 600 feet the creek now plunges off the rim of the basin. It becomes a succession of chutes, cascades and falls for the next mile while it loses over 500 feet of elevation but then almost meanders for a quarter mile across tidal flats near the ghost gold-mining town of Thane to empty into Gastineau Channel. Sheep Creek and valley are thus sharply divided into three parts: a higher, relatively horizontal reach called the upper basin; a middle, vertical jumble of falling white water in a canyon; and a short, rather level, partly tidal coastal section. This physical partition of the watershed is emphasized by corresponding differences in vegetation. The coastal stretch is mostly open, bare or grassy flats; the canyon is densely timbered with big conifers, chiefly Western hemlock, Sitka spruce, and Douglas fir; and the flora of the upper basin and alpine highlands differs utterly from the plants of both lower levels.

Large trees leave off abruptly at the rim of the upper basin—a sudden transition to subarctic vegetation. Only a few spruces grow in the upper basin, but there is a jungle of small willows, alder, salmonberry thickets, and tall weeds, particularly nettle, wild celery, and hellebore, which form a lush subarctic, intermediate floral step to the comparatively insignificant arctic alpine plants of meadows, and the rock, snow and ice fields above. (However, several small "islands" of gigantic cottonwoods thrive near Portal Camp at the confluences of intermittent tributaries with Sheep Creek.) Alpine and arctic influences inevitably mark the upper basin, cooled by vast glaciers. The arctic charr is the characteristic fish, the water ouzel, a typical bird. The mute and shy, though hardy, boreal forms contrast with the spectacular, gross humpy salmon which spawn in the coastal stretch below, their eyes pecked out by the raucous gulls!

The general aspect of the upper basin vegetation strikingly simulates Fishhook Creek plants. The cause is a relatively few rank, abundant, showy species in common. The two widely separated habitats actually share relatively few plants; the two lists have less than ten species in common. The largest and most abundant plants precede in the following list prepared by my wife, Gertrude Frohne. Species she listed (Frohne, 1957) for Fishhook Creek are designated with an asterisk. Characteristic plants of the upper basin, Sheep Creek are:

Alnus sp. (alder), *Salix* spp. (willows), *Rubus spectabilis* (salmonberry), *Populus* sp. (prob. *tricocarpa*), (cottonwood), **Heracleum lanatum* (wild celery), **Epilobium angustifolium* (giant fireweed), *Picea sitchensis* (Sitka spruce), **Veratrum eschscholtzii* (hellebore), *Urtica lyallii* (nettle), **Geranium erianthum* (northern geranium), *Achillea borealis* (northern yarrow), *Aruncus vulgaris* (goatsbeard), *Sambucus racemosa pubens* (elder), *Viburnum edule* (high-bush cranberry), **Aconitum delphinifolium* (monkshood), **Epilobium latifolium* (dwarf fireweed), *Sanguisorba sitchensis* (burnet), *Oplopanax horridus* (devilsclub), *Ribes lacustre* (gooseberry), **Aquilegia formosa*

(columbine), *Rubus stellatus* (nagoonberry), *Aster modestus* (great northern aster), *Rumex arcticus* (dock), *Streptopus amplexifolius,* and *roseus* (twisted stalks), *Actaea arguta* (baneberry), *Impatiens nolitangere* (touch-me-not), *Heuchera glabra* (alpine heuchera), *Tiarella trifoliata* (foam flower), *Claytonia flagellaris* and *siberica* (springbeauty), *Saxifraga punctata* and *tricuspidata* (saxifrages), *Galium triflorum* (bedstraw), *Mimulus guttatus* (monkeyflower), *Campanula rotundifolia* (harebell), *Ranunculus occidentalis* (buttercup), *Cerastium fischerianum* (chickweed), *Geum macrophyllum* (avens), *Circaea alpina* (enchanter's nightshade), *Solidago multiradiata* (northern goldenrod), *Epilobium adenocaulon* (fireweed), *Tellima grandiflora* (fringe-cup), *Prenanthes alata* (rattlesnake root), *Gentiana acuta* (northern gentian), *Veronica tenella* (veronica), *Trifolium hibridum* (alsike clover), *Plantago major* (plantain), *Equisetum arvense* and *hiemale* (horsetail), *Polypodium vulgare occidentalis* (licoricefern), *Cryptogramma acrostichoides* (parsleyfern), *Athyrium ? felix-femina* (ladyfern), *Polystichum ? braunii* (shieldfern).

SUMMARY

The biting rhagionids, *Symphoromyia atripes* and *kincaidi,* were observed and a long series of specimens collected in a mountainous habitat near Juneau, Alaska in an unsuccessful attempt to secure additional specimens of an aberrant form of *kincaidi* of uncertain systematic status known only from this area. The blood-sucking habits and the habitat are described, and the proper assignment of the little known males to species is indicated.

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NORTHEASTERN MOSQUITO CONTROL ASSOCIATION

Host for the 16th annual meeting of the AMCA, to be held at the Somerset Hotel in Boston, Massachusetts on March 27, 28, 29 and 30, 1960.

A REVIEW OF THE MOSQUITO LARVAE OF FRANCE
II. THE GENUS *AEDES*

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Introduction. This paper is the second in a series that is concerned with the identification of the larvae of the mosquitoes known to occur within the geographical boundaries of France. The first paper (Hedeen, 1958) dealt with the genera *Culiseta, Mansonia, Orthopodomyia,* and *Uranotaenia.* A generic key for the mosquitoes found in the country was presented, and the writer gratefully acknowledged the contributions made to this work by his colleagues, both American and French. He wishes to take this opportunity to reiterate his previous statement of thanks to these individuals.

In France there are by far more species in the genus *Aedes* than in any other genus. Due to the fact that the mosquito fauna of the entire European region has not been studied in detail for many years, the modern study of such a large group is very difficult. Many of the species are gradually being synonymized with previously described forms, and a more general realization of the concept of "the species complex" is coming into existence. This is especially true in the cases of the genera *Aedes* and *Culex.* Because of the large number of species of *Aedes* that occur in the country, it was decided to eliminate distributional data which were compiled by the U. S. Army Medical Service during the period of 1953 to 1956 and by other workers. The individuals interested in these data may consult the general references given in the bibliographies of this and the first paper previously cited.

The main objective, therefore, of this particular paper is to present a systematic list of the mosquitoes of the genus *Aedes* reported to occur in France and to give a key by which the fourth instar larvae of these species may be identified. In the preparation of the key it was not possible to examine specimens of a few of the species which were included. In these instances key characteristics were obtained from a study of as many of the previously published descriptions of the species as were possible to secure.

The Present Study. The genus *Aedes* is represented in France by approximately twenty-six species. Twenty-four have been definitely reported, and two are considered by the writer as likely to be present, although not reported at the date of this writing. Several species or varieties described by previous workers have been grouped together as the *Aedes pulchritarsis* complex. It is quite possible that the species included by this writer in the complex will be eventually synonymized with *A. pulchritarsis,* or will be definitely established as independent species when a more detailed study of the group is undertaken. For a detailed discussion of the *pulchritarsis* complex the interested worker is referred to the paper by Aitken (1954, pp. 475–477).

The following species of *Aedes* are considered in this paper. The symbol (*) indicates that larval specimens of that species which had been collected in France were not available for study.

A. Subgenus *Ochlerotatus*
- 1. *pulchritarsis* (Rondani, 1872)
  - a. *berlandi* Seguy, 1921 (*)
  - b. *praeteritus* Seguy, 1923 (*)
  - c. *longitubus* Cambournac, 1948
  - d. *heracleensis* Callot, 1944 (*)
- 2. *rusticus* (Rossi, 1790)
- 3. *refiki* Medjid, 1928
- 4. *excrucians* (Walker, 1856) (*)
- 5. *flavescens* (Muller, 1764)
- 6. *diantaeus* Howard, Dyar, and Knab, 1917

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7. *punctor* (Kirby, 1837)
8. *mariae* (Ed. & Et. Sergent, 1903)
9. *annulipes* (Meigen, 1830)
10. *cantans* (Meigen, 1818)
11. *detritus* (Haliday, 1833)
12. *caspius* (Pallas, 1771)
13. *dorsalis* (Meigen, 1830)
14. *leucomelas* (Meigen, 1804) (*)
15. *communis* (DeGeer, 1776)
16. *pullatus* (Coquillett, 1904)
17. *sticticus* (Meigen, 1838)
18. *nigrinus* (Eckstein, 1918)
19. *cataphylla* Dyar, 1928 (*)
20. *nigripes* (Zetterstedt, 1838) (*)

B. Subgenus *Stegomyia.*
1. *aegypti* (L., 1762)
2. *vittatus* (Bigot, 1861)

C. Subgenus *Finlaya*
1. *echinus* Edwards, 1920 (*)
2. *geniculatus* (Olivier, 1791)

D. Subgenus *Aedes*
1. *cinereus* Meigen, 1818

E. Subgenus *Aedimorphus*
1. *vexans* (Meigen, 1830)

The two species of this genus previously mentioned as not being recorded from France at this time but which are considered likely to occur belong to the subgenus *Ochlerotatus*. They are *leucomelas* (Meigen) and *cataphylla* Dyar. The distribution of *leucomelas* in Europe is compiled by Natvig, 1948. He records it from England, Denmark, Norway, Finland, Sweden, Germany, Austria, Hungary, Poland, and the U.S.S.R. Clavero (1946) reports *leucomelas* from Madrid, Spain. The distribution of *cataphylla* is also compiled by Natvig. He lists this species as occurring in all of the Scandinavian countries, the U.S.S.R., Poland, Hungary, Germany, and probably Austria. Clavero (1946) reports it from Mariana (Cuenca), Spain. Because of the broad distribution of these species from widely scattered areas which are adjacent to the geographical boundaries of France it is considered by the writer that these two species are probably present in the country.

The inclusion of *A.* (*O.*) *nigripes* as a

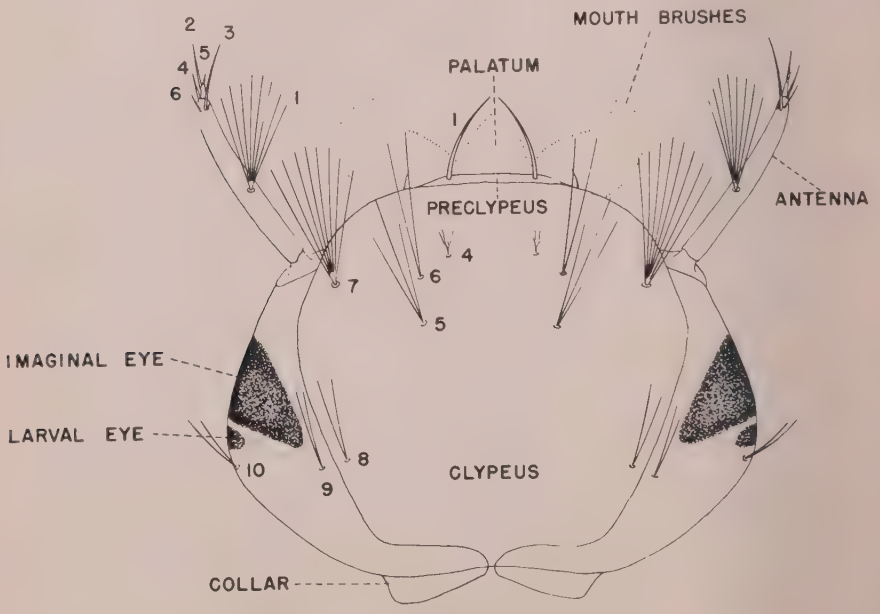

FIG. 1.—Head of *Aedes cantans.*

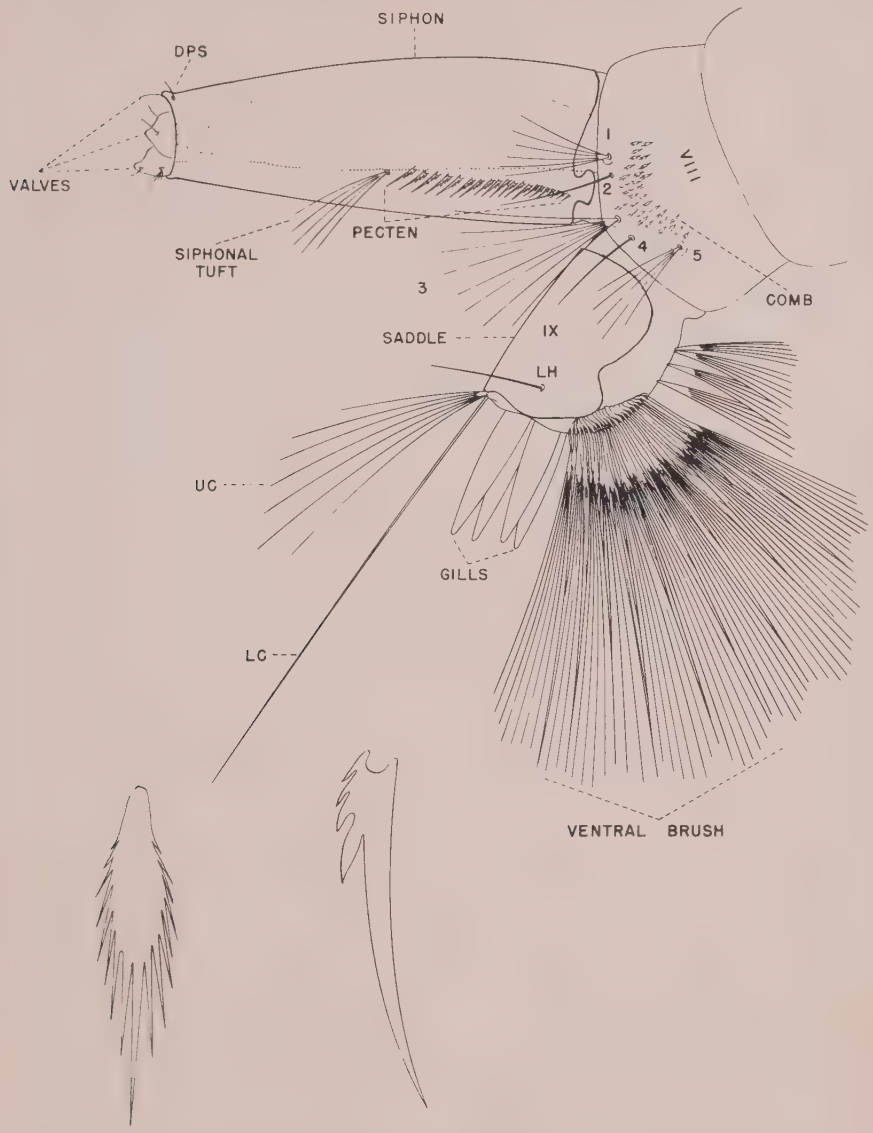

Fig. 2.—Terminal segments of *Aedes cantans*. DPS, dorsal preapical spine. UC, upper caudal. LC, lower caudal. LH, lateral hair of anal segment.

species occurring in France is based on information contained in Seguy (1923, p. 137) and from a personal conversation with Seguy (1955). Seguy includes this species among those listed for the country on the basis of four female specimens in the Paris museum. These specimens are labeled, "Fbl. Juin 69." Edwards (1921) and Natvig (1948) comment that some mistake probably has been made in regard to these specimens. They believe that the labels are incorrect or that the original labels were changed. Seguy, however, remarks that "Fbl." is a common abbreviation for Fountainbleau and that there is little possibility that the labels are incorrect. This writer prefers to adhere to the view of Seguy, and thus includes *nigripes* in the list of *Aedes* occurring in the country.

GENUS *Aedes*

KEY TO FOURTH INSTAR LARVAE

(The morphological features used in this key are labeled in Figures 1 and 2. Variable species are included in the key at several places so that specimens with any of the observed variations may be identified.)

1. Antennal hair (1) single; shaft of antenna not spinose 2
— Antennal hair double or multiple; shaft of antenna with numerous spines and spicules scattered over its surface 4
2. Individual comb scale with a large median spine and several stout submedian spines; length of anal gills usually 2.5 to 4 times greater than length of anal plate, all being of approximately equal size ..... *aegypti*
— Individual comb scale with a large median spine and very small submedian spines; anal gills usually not more than 2x the length of anal plate, two being distinctly smaller than the others 3
3. Pecten extending past middle of siphon; abdominal tufts composed of very long and stout hairs ..... *echinus*
— Pecten not extending past middle of siphon; abdominal tufts composed of shorter and much more slender hairs ..... *geniculatus*
4. Antennal hair (1) two- or three-branched; shaft of antenna usually unspiculated, but sometimes with a few small spines and spicules scattered over its surface 5
— Antennal hair (1) usually much more than two- or three-branched; shaft of antenna more or less uniformly spiculate 6
5. Siphon short, no more than 1.5 to 2 times as long as wide; most distal pecten teeth more widely spaced; subventral tuft inserted within the pecten ..... *vittatus*
— Siphon long, 4 or more times as long as wide; distal pecten teeth not detached; subventral tuft inserted beyond the pecten ..... *pulchritarsis* "complex"
6. Distal few pecten teeth obviously detached or more widely spaced than basal teeth ... 7
— Distal few pecten teeth not obviously detached or more widely spaced than basal teeth 16
7. Anal plate completely surrounding the anal segment ..... *nigripes*
— Anal segment not completely surrounding the anal segment 8
8. Siphon with several hairs along its dorsal surface 9
— Siphon without hairs along its dorsal surface 10
9. Large subventral tuft of siphon inserted within the pecten; anal gills usually not more or less reticulate ..... *rusticus*
— Large subventral tuft of siphon inserted beyond the pecten; anal gills usually presenting a reticulated appearance ..... *refiki*
10. Comb of eighth abdominal segment arranged in a triangular patch 11
— Comb of eighth abdominal segment arranged in an irregular single or double row 14
11. Subventral tuft of siphon inserted within the pecten ..... *cataphylla*
— Subventral tuft of siphon inserted beyond the pecten 12
12. Some comb scales with the submedian spines as long or almost as long as the median spine ..... *caspius*
— All comb scales with a median spine much longer than the submedian spines 13
13. Hair of ventral siphonal valve stout and curved; siphon evenly tapering from base to apex; anal gills usually longer than anal plate ..... *excrucians*
— Hair of ventral siphonal valve slender; siphon distinctly tapering from middle to apex; anal gills usually shorter than anal plate ..... *flavescens*
14. Antennae longer than head ..... *diantaeus*
— Antennae not longer than head 15
15. Upper (5), lower (6), and preantennal (7) head hairs inserted in a line ..... *cinereus*
— Upper (5), lower (6), and preantennal (7) head hairs not inserted in a line ... *vexans*
16. Anal plate completely surrounding the anal segment ..... *punctor*
— Anal plate not completely surrounding the anal segment 17
17. Length of the anal gills less than the length of the anal plate 18
— Length of anal gills greater than the length of the anal plate 25

18. All comb scales with a single outstanding median spine much larger than the submedian spines 19
— Some or all comb scales without a single outstanding median spine much larger than the submedian spines 22
19. Siphon almost as wide distally as basally ..... *mariae*
— Siphon distinctly tapering to apex 20
20. Siphon 3.2 to 4 times as long as broad; lateral hair of the anal segment 1.3 to 1.5 times the length of the anal plate; comb scales varying in number from 20 to 35.. *flavescens*
— Siphon 2.3 to 3.0 times as long as broad; lateral hair of the anal segment 0.6 to 1.0 times the length of the anal plate; comb scales varying in number from 28 to 45.. 21
21. Number of comb scales 31 to 45 ....*annulipes*
— Number of comb scales 28 to 38 .....*cantans*
22. All comb scales uniformly blunt-ended.. *detritus*
— Median and submedian spines of some individual comb scales distinctly larger than median and submedian spines of other individual comb scales 23
23. Subventral tuft inserted beyond middle of siphon .....*caspius*
— Subventral tuft inserted before middle of siphon, rarely inserted at middle 24
24. Anal gills usually globular in shape and no more than one-third (⅓) as long as anal plate; upper and lower head hairs (5 & 6) usually single, either upper or lower not infrequently double .....*dorsalis*
— Anal gills usually distinctly tapering to apex and no more than one-half (½) as long as anal plate; upper and lower head hairs (5 & 6) always single .....*leucomelas*
25. All comb scales without an outstanding median spine 26
— All comb scales with an outstanding median spine 27
26. Upper and lower head hairs (5 & 6) single .....*communis*
— Upper and lower head hairs (5 & 6) double or multiple .....*pullatus*
27. Ventral brush with no more than three pre-cratal tufts 28
— Ventral brush with more than three pre-cratal tufts 29
28. Upper and lower head hairs (5 & 6) almost always double or multiple; comb scales numbering 18–27 (average, 21).. *sticticus*
— Upper and lower head hairs (5 & 6) almost always single; comb scales numbering less than 18 (average, 11) .....*nigrinus*
29. Number of comb scales varying between 31 and 45 .....*annulipes*
— Number of comb scales varying between 28 and 38 .....*cantans*

Summary. This paper is the second in a series concerned with the identification of the larvae of the mosquitoes of France. The genus *Aedes* is considered in this paper, and a key for the identification of the species is included. Several previously described species are grouped into the *pulchritarsis* complex, and consideration is given to species that are considered likely to occur in the country but which have not been reported at this writing.

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NOTES ON THE OVERWINTERING OF *CULEX TRITAENIORHYNCHUS* GILES IN JAPAN[1]

H. R. BULLOCK, W. P. MURDOCH, H. W. FOWLER AND H. R. BRAZZEL

Culex tritaeniorhynchus has been extensively studied in its relation to Japanese encephalitis virus (Mitamura *et al.*, 1938; Petrischeva and Shubladse, 1940; Hammon *et al.*, 1949; Sasa and Sabin, 1950; and others). Taxonomically *C. tritaeniorhynchus* is assigned to the typical subgenus *Culex*, of which all known members overwinter in the adult stage in temperate regions. Sasa (1949) and La Casse and Yamaguti (1950) assumed that *C. tritaeniorhynchus* should conform to the same overwintering pattern and speculated that this species overwinters as adults in small numbers. Precise information on the overwintering habits of this species is of significance in relation to determining whether the virus of Japanese encephalitis overwinters in the arthropod vector as appears to be the case with western equine encephalitis virus in *Culex tarsalis* (Blackmore *et al.*, 1956 and Reeves *et al.*, 1958). Studies on this phase have been pursued by this laboratory for the past eight years, although searches for the overwintering stage of the mosquito have been essentially negative (Anon. 1949–1956).

In the fall of 1957 it was decided to continue the search for overwintering adults primarily because of the observation that this species develops hibernation characteristics when the daylight period becomes less than 12.5 hours and that adult females appear before immature stages and males in the spring. Nakata (1954) observed a single specimen of *C. tritaeniorhynchus* in the laboratory from October 1953 until March 1954. The temperature data presented by Nakata indicate that *C. tritaeniorhynchus* could hibernate under laboratory conditions.

PROCEDURE. Field work commenced with concentration on adult collections from habitats previously unreported as well as from known classical mosquito hibernation habitats. The primary study area was Shinhama in Chiba Perfecture, Honshu, Japan. This area had been previously surveyed for mosquito larvae and adults during the spring and numerous virus isolations had been made during the fall of 1957 from *C. tritaeniorhynchus* by the Department of Virus and Rickettsial Diseases of this organization. Immature stages of *Culex vishnui*, indistinguishable from *C. tritaeniorhynchus* in the adult stage, have not been collected from this area.

Routine dissections of overwintering *C. pipiens* were made to determine the status of hibernation characteristics as have been described by various authors (Swellengrebel, 1929; MacGregor, 1931 and Buxton, 1935). Observations were made on abdominal fat body development, ovarian development, insemination, and evidence of recent engorgement in hibernating mosquitoes to establish criteria for determining whether a *Culex* specimen collected in winter was unequivocally a hibernating adult.

Adult hibernating female mosquitoes were dissected in 0.6 percent saline solution under a dissecting microscope and then the dissected parts were examined under a compound microscope. Sudan IV stain was added to the material on the slide to aid in fat determination. Spermatozoa were readily seen when present by adding slight pressure to the cover slip thus crushing the spermathecae. Subjective categories for fat development were: stage 1 (+++), abdomen with extensive fat body causing distention; stage 2 (++), fat bodies on dissection and abdomen not distended; stage 3 (+), definite fat body development usually found in anterior third of abdomen and traces around sper-

[1] A contribution of the Entomology Department, 406th Medical General Laboratory, APO 343, San Francisco, California.

mathecae; stage 4 (—), few or no fat cells present in abdomen. Subjective categories for ovarian development were: stage 1 (+++), mature eggs; stage 2 (++), yolk (ovum) filling over half the follicle; stage 3 (+), yolk (ovum) filling half the follicle or less; and stage 4 (—), little or no yolk present in follicle. Stages 3 and 4 are comparable to Christopher's (1936) stages 2 and 1 respectively.

RESULTS. The percentages of hibernating characteristics as revealed by dissection of 712 adult *Culex pipiens* collected from October through December 1957 are shown in Table 1. It was observed that all the population of *C. pipiens* did not acquire overwintering characteristics at the same time. Egg rafts of this species were still being collected in October when part of the population had already begun to hibernate. Of approximately 9,000 hibernating *C. pipiens* collected in November through February, none showed evidence of a blood meal. The first spring-collected *C. pipiens* that showed evidence of a blood meal were collected during the second week of March 1958. Because of these data on *C. pipiens* the following combination of characteristics of a winter-collected adult female mosquito is considered to be unequivocal evidence of hibernation and overwintering: (1) presence of fat-body development in the abdomen; (2) lack of ovarian development; (3) presence of spermatozoa in the spermathecae; and (4) lack of recent engorgement, if in the state of true hibernation. Detailed data on hibernation characteristics are presented in the annual professional report of the 406th Medical General Laboratory of 1957 (Anon. 1957).

TABLE 1.—Percentages of hibernation characteristics as revealed by dissections of *Culex pipiens* collected from October through December 1957

Month	Number of specimens	Fat body present			Inseminated	Not engorged	Without ovarian (—) development
		+++	++	+			
October	110	73.6	14.4	03.6	92.7	99.3	99.3
November	333	95.0	00.3	00.7	95.0	99.5	99.5
December	269	96.3	02.6	00.0	98.7	100.0	98.7
Totals	712	91.6	03.4	0.85	96.0	99.8	99.7

During this study, adult *C. tritaeniorhynchus* were collected in the Kanto Plain area every month during winter of 1957–1958, except March. The data on hibernating *C. tritaeniorhynchus* females collected in nature from 18 September through 21 February 1958 are shown in Table 2. Table 2 does not include specimens which were collected in October and were not dissected. All specimens possessed hibernation characteristics when dissected. Although the number of specimens collected was not large, it was established that *C. tritaeniorhynchus* does overwinter as hibernating adults in the Kanto Plain area.

The main hibernating habitats of *C. tritaeniorhynchus* adults were in brush and wood piles of the common pine, *Pinus densiflora* Sieb. et Zucc. Specimens were also collected from bundled bamboo, *Pseudossa japonica* Makino, wood piles composed of *Quercus acuta* Thumb. and *Zelkova serrata* Makino, and a cave. Undoubtedly *C. tritaeniorhynchus* can be collected in other habitats in winter and it is believed that bamboo ground litter should be given particular attention in future surveys. Adults collected from brush piles were often found to be lethargic and did not readily fly when disturbed.

Temperature and humidity data were obtained from a self-recording hygrometer and thermograph apparatus placed 20 inches from the edge and surface of a pine brush pile. The winter temperature ranged from 14° to —1.0° C., but seldom reached 0° C. in winter. It is probable that the centers of large brush piles never freeze.

TABLE 2.—Collection data and results of dissections of hibernating *C. tritaeniorhynchus* females collected in nature from 18 September 1957 through 21 February 1958

	Date	Recent engorgement	Ovarian development	Abdominal fat body	Insemination	Habitat	Location
1	18 Sept. '57	—	—	+	+	pig-baited Magoon trap	Sagiyama
2	18 Sept. '57	—	—	++	+	pig-baited Magoon trap	Sagiyama
3	22 Sept. '57	—	—	+++	+	pig-baited Magoon trap	Shinhama
4	26 Sept. '57	—	—	?	+	pig-baited Magoon trap	Shinhama
5	12 Nov. '57	—	—	+++	+	woodpile *Pinus densiflora*	Kiso near Tokiwa
6	16 Nov. '57	—	—	+++	+	bamboo thicket *Pseudossa japonica*	Shinhama
7	3 Dec. '57	—	—	+++	+	bundled bamboo *Pseudossa japonica*	Shinhama
8	3 Dec. '57	—	—	++	+	brush pile *Pinus densiflora*	Shinhama
9	3 Dec. '57	—	—	++	+	brush pile *Pinus densiflora*	Shinhama
10	18 Dec. '57	—	—	++	+	cut bamboo *Pseudossa japonica*	Shinhama
11	19 Dec. '57	—	—	++	+	woodpile *Zelkova serrata* and *Pinus densiflora*	Onuma near Zama
12	7 Jan. '58	—	—	++	+	brush pile *Pinus densiflora*	Shinhama
13	13 Jan. '58	—	—	+	+	brush pile *Pinus densiflora*	Shinhama
14	14 Jan. '58	—	not checked	not checked	+	brush pile *Pinus densiflora*	Shinhama
15	18 Feb. '58	—	—	+	+	cave	Yokosuka
*16	21 Feb. '58	-	+++	—	+	woodpile *Quercus acuta*	Yokosuka

* This specimen was placed in the insectary on the 21 February 1958. It engorged on rabbit blood on the 26 February and died on 6 March without oviposition. Dissections were performed on the 6 March 1958.

The relative humidity ranged from 76 to 91 percent.

From a total of 11,622 hibernating mosquitoes of all species, collected from all habitats during winter, 4,255 specimens were collected from brush and wood piles.

Russian workers have reported (literature unavailable) that adult females of *C. tritaeniorhynchus* may overwinter in moss, cut grain fields and shrubbery. No specimens have been collected from similar habitats in the Kanto area.

During this study, *Culex vorax* and *Culex rubensis* were collected in winter in Kanagawa Prefecture. This laboratory has no previous record of these two species being collected during winter months.

A few *Culex hayashii* males were collected each month during the winter.

Barr, *vide* Wallis *et al.* (1958), states that after taking a blood meal it seems improbable that *Culex* mosquitoes can undergo the physiological adaptation process (gonadotrophic-dissociation) which is necessary to convert egg production to hibernation. Pilot studies on hibernation characteristics conducted at this laboratory show that it is physiologically possible for colonized *C. tritaeniorhynchus* to develop hibernation characteristics with or without a blood meal when subjected to less than optimal light period.

C. tritaeniorhynchus can be made to develop characteristics, such as fat body development, on a diet of sugar water when subjected to less than an optimum light schedule. If this occurs in nature, it would be unlikely that this species would be an overwintering host for virus, unless transovarian transmission is involved. Perhaps there is not a true gonotrophic dissociation in *C. tritaeniorhynchus* as stated by Newson and Blakeslee (1957) but just the lack of a blood meal stimulus for ovarian development. It may be possible to determine whether a hibernating *Culex* mosquito takes a blood meal before hibernating by determining nitrogen levels in experimentally fed mosquitoes over different time intervals and in mosquitoes collected in nature throughout the year, and particularly in the winter.

Other observations on hibernating characteristics were that ovaries of laboratory reared *C. tritaeniorhynchus,* fed only on sugar water, occasionally develop to the stage in which the ovum occupies half of the follicle. When a group of *C. tritaeniorhynchus* females are given a single blood meal, fresh apples, and subjected to less than optimal light period, part of the group will develop overwintering characteristics such as gonadotrophic dissociation. This was accomplished experimentally by placing caged and recently engorged mosquitoes in a location where they did not receive the benefit of a light control box that extended the day light period to 15 hours.

Summary. During the winter of 1957–1958, it was determined that *C. tritaeniorhynchus* overwinters as adults in Japan. The main habitats in which specimens were collected were brush and wood piles of the common pine, *Pinus densiflora.* Specimens have also been collected from wood piles of *Zelkova serrata* and *Quercus acuta,* bundled bamboo, and from a cave. Data from pilot laboratory studies indicate that there is a possibility that *C. tritaeniorhynchus* does not require a blood meal to overwinter. If this be the case in nature, it would be unlikely that this species would be an overwintering host for virus unless transovarian transmission is involved.

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NEW RECORDS AND DISTRIBUTIONS OF THE BITING FLIES OF MT. DESERT ISLAND, MAINE [1]

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During 1955 and 1956 the author spent parts of the summers on Mt. Desert Island. During these visits he made extensive collections of the insects with the purpose of continuing the survey, initiated by Dr. William Procter, of the fauna of the Island.

The present paper considers the biting flies which were given particular attention for several reasons. The importance of many of these insects as pests, together with the ability of certain ones to act as disease vectors, makes it essential that we know what species are present, when they occur and where they are abundant. In addition the author wished to obtain specimens for use in classes.

The biting flies which were collected included five families, the Simuliidae, the Culicidae, the Heleidae, the Tabanidae and the Muscidae.

The family Simuliidae was not particularly abundant during either year. This undoubtedly is related to the time of the year since these insects are reported to be common and troublesome on the Island in May. In 1955 two specimens of *Prosimulium hirtipes* (Fries) represented the total catch. These were collected on the trail to Beach Cliff on July 9. In 1956 specimens of *Simulium venustum* Say were collected from the following localities: Oak Hill (8) VI-4–56. Seawall (5) VI-5–56. Seal Harbor (6) VI-5–56. Western Mountain (5) VI-4–55. *Prosimulium hirtipes* (Fries) was recorded from the following localities: Beach Cliff (2) VI-4–56. Oak Hill (3) VI-4–56, Seal Cove (5) VI-5–56. Seawall (2) VI-5–56. Seal Harbor (7) VI-5–56. Western Mountain (6) VI-4–56.

The family Heleidae, while present in some numbers, was not found to be rich in species. In 1955, a single species *Culicoides canithorax* Hoff. was a common and troublesome pest about dusk at Seawall and also at Ship Harbor. In 1956, in addition to a large series (187) of *C. canithorax,* a few specimens of *C. melleus* (Coq.) and *C. obsoletus* (Meig.) were taken.

The family Culicidae was well represented. Five genera including 18 species were secured. In 1955 the emphasis was placed on adult collections, while in 1956 the survey was continued primarily by larval dippings. Their distribution is indicated in the accompanying summary.

Aedes: abserratus (F. & Y.) Seawall (1) VII-8–55. Seawall Bog (110) VI-3–56; — *atropalpus* (Coq.) Thunder Hole (5) VII-1–55; (24) VII-12–55; (175) VI-3–56; (2) V-12–56; (2) X-22–56. Ship Harbor (2) VII-19–55. Seawall (1) VII-13–55. Anemone Cove (6) VII-1–55; (30) VII-12–56; (25) VII-25–56; (4) X-2–56. Otter Cliff (10) VI-30–56; (40) VII-25–56. Bass Harbor (30) VII-25–56. Sand Beach vicinity (60) VI-3–56; — *aurifer* (Coq.) Seawall (1) VII-9–55; — *canadensis* (Theo.) Ship Harbor (4) VII-2, 10–55. Seawall (2) VII-8, 10–55; (70) VI-4–56. Echo Lake (4) VI-4–56. Seal Harbor (20) VI-5–56. Cadillac Mt. (50) VI-6–56; — *cantator* (Coq.) Seawall (29) VII-3, 18–55. Ship Harbor (10) VII-2, 10, 11–55. Tremont (300) VII-2, 27–55; (90) VI-3–56. Sand Beach (10) VI-4–56. Bernard (50) VI-6–56; — *cinereus* Meig. Seawall (6) VI-3–56. Echo Lake (4) VI-4–56; — *communis* (Deg.) Seawall (3) VII-3, 9–55; — *decticus* H. D. & K. Seawall (1) VII-8–55; — *diantaeus* H. D. & K. Seawall

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(2) VII-4, 6–55; — *excrucians* (Walk.) Tremont (1) VII-2–55. Seawall (3) VII-2, 8, 13–55; (30) VI-4–56. Otter Creek (5) VI-6–56; — *fitchii* (F. & Y.) Seawall (1) VII-18–55; (2) VI-3–56; — *intrudens* Dyar Ship Harbor (1) VII-10–55; — *punctor* (Kby.) Seawall (15) VII-3, 9–55; (23) VI-3–56. Tremont (58) VI-5–56. Echo Lake (45) VI-4–56; — *sollicitans* (Wlk.) Seawall (2) VII-9, 18–55. Ship Harbor (9) VII-2, 19–55. Tremont (12) VII-2, 27–55. Bernard (5) VII-25–56.

Culex restuans (Theo.) Sand Beach vicinity (5) VII-25–56.

Culiseta morsitans (Theo.) Seawall (7) VI-3–56.

Mansonia perturbans (Wlk.) Ship Harbor (8) VII-3–55. Tremont (3) VII-2, 4, 27–55.

Wyeomyia smithii (Coq.) Seawall VII-4–55. Seal Cove Pond VII-6–55. Common in *Sarracenia purpurea.*

The family Tabanidae was abundant both in species and numbers. Five genera including 22 species were obtained. The records of their distribution follow.

Chrysops: carbonaria (Wlk.) Bernard (1) VII-2–56; — *celeris* O.S. Ship Harbor (1) VII-2–55; — *excitans* (Wlk.) Cadillac Mt. (1) VII-9–55. Great Heath (3) VII-8–55. Parkman Mt. (4) VII-12–55. Seawall (1) VII-24–55. Ship Harbor (2) VII-2, 15–55. Somesville (2) VII-1–55. Acadia Mt. (1) VII-15–56; — *frigida* O.S. Great Heath (1) VII-8–55. Ship Harbor (1) VII-15–55. Seawall (1) VII-26–56; — *fuliginosa* (Wied.) Ship Harbor (1) VII-2–55. Tremont (1) VII-2–55; — *lateralis* (Wied.) Western Mt. (1) VII-6–55; — *mitis* O.S. Duck Pond (1) VII-5–55. Seawall (1) VII-4–55; — *montana* O.S. Carter's Nubble (1) VII-10–55; — *nigra* (Macq.) Ship Harbor (2) VII-1–55. Somesville (1) VII-2–55. Tremont (1) VII-2–55; (1) VII-2–56; — *vittata* (Wied.) Carter's Nubble (1) VII-10–55. Otter Creek (1) VII-17–55. Western Mt. (1) VII-6–55.

Atylotus thoracicus (Hine) Ship Harbor (1) VII-20–56. Seawall (1) VII-26–56. Great Head (1) VII-27–56.

Hybomitra: affinis (Kirby) Cadillac Mt. (1) VII-9–55; — *aurilimba* (Stone) Sargent Mt. (1) VII-16–56; — *frontalis* (Wlk.) Sargent Mt. (5) VII-26–55. Tremont (4) VII-13–55; (1) VII-11–56; — *illota* (O.S.) Tremont (2) VII-11–56. Seawall (1) VII-26–56. Great Head (1) VII-27–56; — *minusculus* (Hine) Duck Pond (2) VII-3–55; — *trispila* (Wied) Sargent Mt. (1) VII-26–55. Seawall (1) VII-28–56; — *zonalis* (Kirby) Seawall (1) VI-30–55.

Merycomyia whitneyi (Johns.) Sargent Mt. (1) VII-26–55.

Tabanus: lineola scutellaris (Wlk.) Tremont (1) VII-29–55; — *nigrovittatus* Macq. Seawall (2) VII-24–55. Tremont (5) VII-27–55; (1) VII-11–56; — *nivosus* (O.S.) Seawall (1) VII-24–55.

Two species of biting muscid flies were taken. The horn fly *Siphona irritans* (L.) was observed in Somesville and on the road to Beach Cliff. *Stomoxys calcitrans* L. was a common and annoying pest in many locations along the shore.

SUMMARY

Data are presented concerning the prevalence and occurrence of biting flies collected during the summers of 1955 and 1956 on Mt. Desert Island.

Blackflies (Simuliidae) represented by two genera were not particularly abundant or troublesome although they are recorded to be common and annoying earlier in the season.

The family Heleidae was represented by *Culicoides canithorax, C. melleus* and *C. obsoletus* with *canithorax* being the most abundant. Of these *C. melleus* and *C. canithorax* have not been recorded previously from Mt. Desert (Procter, 1946). *C. obsoletus* has been reported from Long Pond as *C. ranguisuga* (Coq.).

The Culicidae included five genera and 18 species. Of these *Aedes abserratus, A.*

decticus and *Culiseta morsitans* had not been reported from the area according to Procter 1946. *Aedes atropalpus* previously reported from Bar Harbor was found to occur quite commonly wherever there are rocky ledges extending above the high tide level.

The Tabanidae are represented by 22 species and five genera. *Chrysops montana, Hybomitra frontalis, Merycomyia whitneyi* and *Tabanus nivosus* are new records both for Mt. Desert Island and for Maine. *Hybomitra aurilimba* is a new record for the Island.

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NOTES ON THE TECHNIQUES OF HANDLING MOSQUITOES IN THE LABORATORY [1]

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In the laboratory, techniques are often a matter of personal preference. However, a communication of this nature may nevertheless be very useful. For extensive information on mosquito culture techniques and experimental procedures, Trembley's paper (1955) should be consulted. Reported here are some experiments and techniques which either are not covered by Trembley or are considered improvements by the author.

Cage. For the conventional screen-type of mosquito cage, nylon is the most desirable, because it is lighter than wire, more durable than cheesecloth, and above all, offers better visibility than either. Since adequate humidity is essential for the survival of the adult mosquitoes, ordinarily it is supplied to this type of cage by providing wet toweling or by putting the cage in a specially humidified room. Frequently the cage becomes moldy and difficult to clean.

In our laboratory, cages with favorable humidity but with no mold contamination have been maintained in their original condition for the past five years.

The cage is very simply constructed. It is composed of a plywood top and bottom, four corner bars, and an extra horizontal bar to square off a lower opening for the nylon sleeve. The rest of the framed area is walled with "Clear Acetate" sheet, a plastic that is transparent, durable, non-inflammable and easily cleaned by a damp sponge.

Moisture inside the cage is supplied from a piece of cheesecloth drawn out from a water bottle. The cloth is spread and hung over a glass rod resting on two hooks screwed on the corner bars of the cage. In such a closed cage, the relative humidity

[1] This investigation was aided in part by a grant E-87 to Dr. G. H. Ball, from the National Institutes of Health, U. S. Public Health Service, and by a Grant 254 from the Board of Research, University of California.

can be kept above 60 percent with one piece of cheesecloth, and above 80 percent with two pieces, one on each side of the cage. For efficient transpiration, distilled water should be used in the bottle and the cloth should be bleached or replaced when it becomes stiff.

The wood frame is painted with enamel paint, and hence the cage can be cleaned easily by wiping with a damp cloth or washing, and kept in top condition at all times.

ADULT TRAPPING TUBES. For general routine in our laboratory, a suction tube attached to the compressed air line was used instead of a mouth suction tube. In picking up adults for experimental purpose, a simple glass trapping tube was employed.

This tube had a bore of 1 cm. and a length of about 45 cm., depending upon the size of the cage in which it was to be used. It was slightly flared at one end over a flame. About 6 cm. from this end it was bent broadly to an angle of about 112°.

For trapping mosquitoes, the handling end of the tube was plugged with cotton and the flared end was brought directly over the mosquito. This was quickly lowered onto the mosquito, which in trying to take flight, was trapped at the bent section of the tube. A gentle tapping sent this mosquito to the other end, and the tube was ready to catch the next one.

After a little practice three or four mosquitoes could be caught in a minute. It was found convenient to have many such tubes made, and thus mosquitoes trapped in small lots of 5 or 6 could be handled more easily for experimentation. This method was especially good for picking up engorged females which might be injured by suction.

ANAESTHESIA. Carbon dioxide gas was preferred to ether or chloroform because it could be easily passed through the trapping tube, and if it was applied to a closed chamber such as lamp chimney, it did not have an area of high concentration that could kill the mosquitoes falling close by.

DE-WINGED ADULTS. In our laboratory, it was found that *Culex tarsalis* and *C. pipiens* lived a normal length of life after their wings were pulled out at the base. With two pairs of forceps, such an operation could easily be performed while the insects were still under the influence of CO_2. Judged from the mortality immediately following the operation and thereafter, the mosquitoes apparently were not harmed. If they were supplied with food, they could be raised as walking insects in Petri dishes.

Kept as de-winged adults, *C. tarsalis* females laid viable eggs as usual after a blood meal on a malarial canary. Likewise, the sporogonous development of *Plasmodium relictum* in the mosquito was completed in a normal manner. We have found that such a technique certainly streamlined our dissection work, and we hope it will be found useful by fellow "mosquiteers."

EGG COUNTS. It is impractical, if not impossible to count the eggs while they are floating on the surface of water. In egg-hatching studies on *Culex* mosquitoes in our laboratory, the egg cases were submerged in a thin layer of 70 percent alcohol and the eggs were counted under a dissection microscope, after the hatching was finished.

If great accuracy is not demanded, the percentage of hatching can be determined by counting only the eggs in the peripheral layer of the raft. By this method, counts can be done under the dissection microscope while the rafts are still floating on the surface of water without being teased apart. The hatched and unhatched eggs are very apparent in the unbroken raft.

In insects which lay circular or near circular egg masses, this counting method is very accurate. One may choose to count the eggs along the diameter or around the circumference in an egg mass to get values both for the total number of eggs and the hatching ratio.

MOSQUITOES IN THE DARK ROOM AT 18° C. *C. tarsalis* and *C. pipiens* went through many generations in a dark room at a constant temperature of 18° C. although the populations and life processes

(biting, egg laying and hatching, length of each life stage) were subnormal. Mortality in the first larval instar was high, but emergence was normal and the pupae and adults were larger than those at room temperature (above 26° C.).

Our *C. pipiens* is an autogenous strain, and the autogenous character is demonstrable under the above mentioned conditions. Our *C. tarsalis* is capable of laying viable eggs without a blood meal at room temperature (Chao, 1958) but a blood meal is necessary for viable eggs in the dark at 18° C.

In answer to a mosquito control problem, Shute (1959) pointed out that *Culex molestus* bred autogenically in air raid shelters in London during the last world war. Earlier, Jobling (1937) reported that *C. pipiens, C. fatigans,* and *Aedes argenteus* could develop in complete darkness, and a second generation was produced by the autogenous *C. pipiens*. It is evident that some autogenous strains of mosquito can reproduce in the absence of light, a fact that should be taken into consideration in mosquito eradication programs.

Experiments in Hybridization of *C. tarsalis* and *C. stigmatosoma*. Since our laboratory had both colonies of *C. tarsalis* and *C. stigmatosoma* in 1955 (Ball and Chao, 1956) several attempts were made at that time to cross them. The results of the trials were negative regardless of in which direction the cross was made. No sperm was found in the spermacecae of the females of either species. Egg cases were laid after a blood meal but the eggs were not viable.

Summary. Reported in this paper are an improved mosquito cage, techniques of catching and de-winging of the adults, and the method of egg counting. The raising of mosquitoes in a dark room at reduced temperature is reported. Attempts to hybridize *Culex tarsalis* and *C. stigmatosoma* were unsuccessful.

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UTAH MOSQUITO ABATEMENT ASSOCIATION

Sixty per cent of the people in the state of Utah are now living within the boundaries of organized mosquito abatement districts.

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Proceedings of Annual Meetings for Sale.

A SURVEY OF THE MOSQUITOES OF THE SOUTHERN RYUKYUS

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University of California, Davis

Requests for information on the mosquitoes of the Ryukyuan area, and recent activity among Japanese workers, have prompted the assembling and publication of the data presented below. These notes are based mainly on an unpublished mimeographed report submitted to Ryukyuan authorities in 1951. Since that time the identifications have been verified with the help of Dr. Alan Stone of the U.S. National Museum, and Dr. K. L. Knight of the U.S. Navy. Assistance during the period of collection was rendered by Dr. Shinken Ohama and Mr. Koubo Miyara.

Mosquito larvae and adults were collected during a three-month period, October to December, 1951 on the islands of Ishigaki and Iriomote, the two largest islands of the southern Ryukyus, known in the aggregate as Yaeyama. Several undescribed species and many new records resulted.

The study was made under the auspices of the SIRI Program, Pacific Science Board, National Research Council, while working with the Public Health and Welfare Department, United States Civil Administration of the Ryukyu Islands.

The collection period of October to December was probably not so favorable from the standpoint of quantity of material as midsummer would have been. Weather conditions were often poor—cloudy, windy and rainy—making field work difficult if not impossible. Temperatures fell abruptly toward the end of November, with a generally depressing effect on insect populations. Nevertheless, the 38 species of mosquitoes collected are considered representative, especially since a survey by Teller and Gentry (1955) made during January, 1954 revealed no additional species and a total of only 18. However, it must be said that January is an unusually poor month for field investigations.

Ishigaki Island is extensively mountainous but the highest elevation is reached by Mt. Omoto at 1,750 feet. Along the coast and in the southern half of the island there is considerable flat and sloping land suitable for agriculture. The important crops are sweet potato and rice with much smaller acreages devoted to soybeans, peanuts and sugarcane. The only large river is the Miyara, which nearly bisects the island. However, there are about 35 other small rivers, some of which may be of considerable importance with respect to malaria. The mountains are heavily forested, but mostly with small, second growth timber. Tree holes, which harbor several types of mosquito larvae in addition to *Culicoides,* are common only where species of *Ficus* occur. Large numbers of these are present in two collecting sites, along the Tori River and on the Yarabu Peninsula.

Iriomote Island is the largest of the Ryukyus south of Okinawa. It is almost completely covered with low but rugged mountains and is traversed by six large rivers as well as many smaller ones. Agriculture is confined to the relatively flat areas along the coast and adjacent to the larger rivers. Here, also, the culture is one of sweet potato and rice.

The climate of Yaeyama is similar to that of Okinawa with warm weather from April to November and winter temperatures in the low forties. The rainfall averages 86.4 inches on Ishigaki and 109.3 inches on Iriomote. September is the rainest month but there is enough rainfall distributed over the year to maintain a large population of insects.

An effort was made to collect in all types of environments such as rice paddies, road puddles, drying creek bottoms, stream eddies, rock holes, tree holes, leaf axils, artificial containers, crab holes, polluted pools, fresh and brackish water. No water was found in epiphytes, and no pitcher-plants were located. Furthermore, no mos-

quito larvae were observed in temporary rain puddles nor in brackish water. This does not eliminate any of the above as possible breeding sites, however. Collections were made in forest, jungle, grassland, swamp, cultivated and domestic areas. In addition to the many larvae taken, several hundred adults were netted in underbrush and near damp rocks. Species collected only as biting females were *Mansonia uniformis* (Theobald), *Mansonia crassipes* (v. d. Wulp), *Aedes* (*Aedes*) sp., and *Aedes albocinctus* Barraud.

The total collection consisted of 1,234 pinned adults, about 500 slide-mounted larvae, and over 1,000 larvae in alcohol.

Of the 38 species of mosquitoes located, 33 were found on Ishigaki and 31 during a 3-week November stay on northern Iriomote. This compares with 37 species in the much more intensively collected island of Okinawa. The total for the three islands is 49. The species are obviously a part of the Oriental fauna and many of them are widespread in that region. However, 7 of the 37 Yaeyama species are unknown outside the Ryukyus. Most of the others were new records for the southern Ryukyus.

Useful publications bearing on mosquitoes of the area are Barraud (1934), Bohart and Ingram (1946), LaCasse and Yamaguti (1950), Teller and Gentry (1955), and Bohart (1956).

The following list of species includes notes on distribution, ecology, abundance, and evidence of man-biting habits.

Anopheles minimus Theobald. Widely distributed in the Oriental Region. Larvae are usually found in the eddies or grassy edges of clear, fast-flowing, sunlit streams. It is a prime malaria vector but fortunately is much less common than *sinensis*. On Ishigaki it is most abundant along the north coast, the northern peninsula, and Fanan River where I collected many specimens. On Iriomote it is reported from the more swiftly flowing parts of most of the rivers as they approach the coast.

Anopheles ohamai Ohama (1947). This species breeds with *minimus* and could be a malaria vector if it were more common. It is known on both islands. I collected it in the Fanan River of northeastern Ishigaki. It is closely related to *saperoi* Bohart and Ingram from Okinawa but *ohamai* has the pale costal spot beyond the wing middle about as large as the subcostal spot, instead of smaller. In the larva the postclypeal hair is shorter.

Anopheles sinensis Wiedemann. A common rice-paddy species throughout the Orient. It also frequents grassy spring pools and other semi-stagnant places. It bites at night but is somewhat zoophilic. It is probably responsible for some malaria each year but is far outranked as a vector by *minimus* during periods of population movement as in war-time.

Toxorhynchites yaeyamae Bohart. A rare tree-hole species. Four larvae were captured, one to a hole. Three of these came from Shirahama, Iriomote and one from the Yarabu Peninsula, Ishigaki. A similar but unnamed species occurs on Okinawa and still another, *yamadai* Oûchi, inhabits the northern Ryukyus. The larval, pupal and adult descriptions of *yaeyamae* were given by Bohart (1956).

Tripteroides bambusa Yamada. The common "fuzzy" larva of *Ficus* tree holes on both islands. The species is well known in Japan but does not seem to occur on Okinawa.

Ficalbia luzonensis (Ludlow). One adult of this widespread Oriental species was taken in shrubbery near Hoshitate, Iriomote.

Harpagomyia genurostris (Leicester). A widespread Oriental species breeding in leaf axils of large taro plants. It was common on both islands.

Uranotaenia annandalei Barraud. Known from India and China, it is rare on Okinawa, but abundant on Ishigaki and Iriomote. The larval antenna is unusual in having 3 leaf-like appendages. The favored habitat is a drying creek pool. Ishigaki localities were Kawara, Mt. Banna, Mt. Maeshi, Kabira, and Fanan River. Iriomote collections were made near Sonai, the Nakara River, and Shirahama.

Uranotaenia bimaculata Leicester. A tree-hole species widespread in the Orient. I found it uncommonly in tree holes on Mt. Omoto and Mt. Maeshi, Ishigaki, and swept it from shrubbery along the Nakara River, Iriomote.

Uranotaenia macfarlanei Edwards. Another widely distributed Oriental species which was fairly common in Yaeyama. On Ishigaki it was taken on Mt. Kawara, Mt. Banna, and Kabira. On Iriomote it was collected near Sonai and along the Nakara River. The larvae inhabit small ground pools.

Uranotaenia stonei Bohart. An endemic Ryukyuan species found originally in northern Okinawa. It is common on Ishigaki where larvae breed in cave springs, rock holes and crab holes. Larvae are of the sinuous *"Aedes"* type, contrasting with the *Anopheles*-like other Yaeyama species. Adults were disturbed from shrubbery on Mt. Banna, Mt. Kawara, Mt. Maeshi, and the Yarabu Peninsula of Ishigaki.

Armigeres subalbatus (Coquillett). A well-known Oriental species, widely scattered in Yaeyama but uncommon. Adults bite readily by day. Larvae were bred from tree holes along the Tori River and from taro axils along the Fanan River, Ishigaki.

Orthopodomyia anopheloides (Giles). Not previously recorded from the Ryukyus, this Oriental species is common in tree holes on Ishigaki and Iriomote.

Mansonia crassipes (v. d. Wulp). This reddish species was taken in shrubbery rather frequently on Iriomote from Shirahama to Hoshitate.

Mansonia uniformis (Theobald). Adults of this ubiquitous Oriental species were collected several times day-biting and occasionally sweeping. Localities were Yarabu Peninsula on Ishigaki, and Sonai, Hoshitate and Shirahama on Iriomote.

Aedes (*Aedes*) *ishigakiensis* Bohart. A long series of adults were reared from a foxhole in a pine grove on Mt. Banna, Ishigaki. Larval, pupal and adult characters were given by Bohart (1956).

Aedes (*Aedes*) sp. Two females of an unidentified species near *atrius* Barraud were taken day-biting in the forest east of Hoshitate, Iriomote.

Aedes (*Finlaya*) *albocinctus* Barraud. Three females of this Oriental species were taken day-biting on Iriomote, two on the mountain back of Shirahama, and one above the east fork of the Nakara River.

Aedes (*Finlaya*) *feegradei* Barraud. This Burmese species was reported previously on Okinawa by Bohart (1953). Adults were reared from larvae taken in deep and narrow *Ficus* tree holes near the Yarabu River, Ishigaki.

Aedes (*Finlaya*) *japonicus* (Theobald). This Japanese species is common in rock holes along streams on both islands.

Aedes (*Finlaya*) *okinawanus* Bohart. Previously known only from Okinawa, this species occurs sparingly in Yaeyama. Adults sometimes day-bite in deep shade. It breeds in tree holes and was taken on Mt. Omoto and Yarabu Peninsula, Ishigaki, as well as Sonai, Shirahama and Nakara River, Iriomote.

Aedes (*Stegomyia*) *aegypti* (Linnaeus). Found practically around the world, this species was common in artificial containers of villages along the south coast of Ishigaki. It is a well-known vector of dengue in the Pacific area along with the following species.

Aedes (*Stegomyia*) *albopictus* (Skuse). A widespread Oriental species which was found breeding in small tree holes on Mt. Maeshi and in a metal drum at Shiraho, Ishigaki. Adults were day-biters but not numerous enough to be pestiferous.

Aedes (*Stegomyia*) *flavopictus downsi* Bohart. This mosquito is a dominant day-biting pest in wooded areas. It breeds frequently in taro axils and sometimes in tree holes. It is known only from Okinawa and Yaeyama, where it is common on both Ishigaki and Iriomote.

Aedes (*Stegomyia*) *riversi* Bohart. Second only to *downsi* as a woodland day-biter. It is abundant on both islands where the larvae breed primarily in tree holes. As the larvae were not found in rock holes, this may be a subspecies of the Okinawan form.

Culex (*Lutzia*) *vorax* Edwards. The larva is predaceous in ground pools and artificial containers. Adults bite man. It is widespread on both islands, but not common.

Culex (*Lophoceraomyia*) *cinctellus* Edwards. An Oriental species often swept from shrubbery in Yaeyama. Larvae breed in open grassy ground pools.

Culex (*Lophoceraomyia infantulus* Edwards. A well-known Oriental species which is widespread but uncommon in Yaeyama. The larvae breed in ground pools.

Culex (*Lophoceraomyia*) *minor* Leicester. This Oriental species is a common tree-hole and rock-hole breeder everywhere in Yaeyama.

Culex (*Neoculex*) *hayashii* Yamada. Not uncommonly breeding in springs and cutoff stream pools on Ishigaki but not found on Iriomote. Elsewhere it is known from Japan, China, and Okinawa.

Culex (*Culiciomyia*) *nigropunctatus* Edwards. Not previously reported from the Ryukyus but occurring over much of the Orient. The larva has a peculiar false joint on the siphon. It was found in grassy ground pools on Mt. Banna, Ishigaki, and along the Nakara River, Iriomote.

Culex (*Culiciomyia*) *ryukyensis* Bohart. Collected once in a ground pool on Mt. Banna, Ishigaki, and once in a cutoff stream pool near Shirahama, Iriomote. This species is common in northern Okinawa.

Culex (*Culex*) *bitaeniorhynchus* Giles. Generally found in rice paddies and algal ground pools on both islands as well as elsewhere in the Orient. It is a night-biter but not abundant enough to be much of a pest.

Culex (*Culex*) *quinquefasciatus* Say. This worldwide species breeds in almost any form of stagnant water, especially if polluted with sewage or food. In Yaeyama, larvae are common in laundry sumps, clogged sewer drains, and artificial containers. Its role in transmission of filariasis is well known.

Culex (*Culex*) *sinensis* Theobald. A rice-paddy breeder throughout the Orient and probably well distributed in Yaeyama. However, I took only one adult from shrubbery on Mt. Maeshi, Ishigaki.

Culex (*Culex*) *tritaeniorhynchus* Giles. This prolific Oriental mosquito was infrequently found in Yaeyama, but is common on Okinawa. It is a ground pool breeder and incriminated in transmission of Japanese "B" encephalitis in Japan and Okinawa.

Culex (*Culex*) *vishnui* Theobald. A widespread Oriental species but scarce in Yaeyama. I collected larvae once in a swamp above a rice paddy near Kabira, Ishigaki, and once in a grassy pool along the Nakara River, Iriomote. The adult bites man.

Culex (*Culex*) *whitmorei* (Giles). Essentially a rice-paddy breeder throughout the Orient, but not previously recorded from the Ryukyus. Larvae were found near Sonai, Iriomote. Adults are known to bite man.

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OPERATIONAL AND SCIENTIFIC NOTES

DISCOVERY OF *Mochlonyx fuliginosus* (Felt) IN PASSAIC COUNTY, NEW JERSEY.—This rare species of *Mochlonyx* was collected by the author in the larval, pupal and adult stages in West Milford Township, Passaic County, New Jersey. No records have been found of previous collections of this species in New Jersey.

The breeding site is a cold sphagnum bog heavily shaded around its perimeter by hemlock (*Tsuga canadensis*) white pine (*Pinus strobus*) and rhododendron (*R. maximum*) with a thick growth of *Vaccinium* spp., swamp azalea (*Rhododendron viscosum*) swamp laurel (*Kalmia polifolia*) and many herbaceous plants growing in the water and upon the hummocks.

Observations throughout six or seven years in the particular area of the bog where *M. fuliginosus* were collected have shown that during the summer all standing water disappears but the sphagnum and leaf litter remain quite damp. After heavy late summer and fall rains and subsequent raising of the water table the bog becomes flooded and remains so until the following June or July.

All specimens of *M. fuliginosus* were collected in late summer and fall and one adult was even found emerging from the pupal skin on December 1, 1955 while a skim of ice was present on the breeding site. Larvae were collected September 11, 1952 and pupae were found on September 15, 1955 and September 27, 1957.

Although the life history of this species has not been established it seems possible, since adults have been collected in May, June, July and August in other states, that some eggs laid by these early emerging females hatch with the late summer and fall rains producing the late broods such as found in this breeding site, in Passaic County, New Jersey.

All *M. fuliginosus* specimens, including male genitalia slides, were sent to Professor Edwin F. Cook of the Department of Entomology and Economic Zoology, University of Minnesota for confirmation. Professor Cook was especially interested in examining larvae and pupae which were unknown up to this time. He will report on these elsewhere.—Robert W. Lake, Assistant Superintendent, Passaic County Mosquito Extermination Commission.

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A NOTE ON *Mansonia* BREEDING IN OREGON LOG PONDS.—During 1956, cooperative studies on the ecology and control of log pond mosquitoes were carried out at Roseburg, Oregon, by the Logan Field Station Section of the U. S. Public Health Service, the Oregon State Board of Health, and the Douglas County Health Department. In the course of these investigations, a major breeding site for *Mansonia perturbans* was discovered in a group of log ponds near Sutherlin, Oregon.

The ponds studied range in area from 10 to 20 acres, and are about 10 feet deep. The logs are stored in bundles 8–10 feet in diameter, held together by steel bands.

During the 10 years that the ponds have been in use, the margins have become occupied by a dense stand of cattail (*Typha* sp.), which has extended out from the banks for 10 feet or more as a floating mat. Parts of the mat have become detached, forming floating islands. Cattail also grows abundantly on the emergent tops of the floating log bundles, the roots extending down through the logs into the water.

During surveys in December and late May, large numbers of *Mansonia perturbans* larvae were found on the roots and rootstocks of the cattail plants. During the late July and early September surveys few larvae could be found attached to the plants, although they were readily obtained by scraping the under side of floating cattail mats with a long-handled pint dipper. This seasonal difference in larval behavior is probably due to the low water temperatures prevailing in winter and spring, the larvae being then too lethargic to release rapidly.

May samples contained only 4th instar larvae and pupae, July samples 2nd and 3rd instar larvae, and the September and December samples 3rd and 4th instar larvae. These observations substantiate that there is a single generation each year (Horsfall, 1955), that the species overwinters as larvae, and that emergence takes place in early summer. A light trap installed near the ponds showed *Mansonia perturbans* to be the most abundant mosquito species in the area in June and July.

It is suggested that surveys of *Mansonia* breeding areas be carried out in cold weather, as the larvae are then easily obtained by pulling up plants. Another large wetland plant, *Juncus effusus,* grows in the water at the edge of the ponds in association with cattail; no larvae were ever found on *Juncus* roots, probably because they are woody in consistency and lacking in large air spaces.—Laverne S. Miller, Oregon State Board of Health, Portland, and Robert A. McHugh, C.D.C., P.H.S., Greeley, Colorado.

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EDITORIAL

WILDLIFE FROM THE MOSQUITO CONTROL VIEWPOINT [1]

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In the execution of a mosquito control program, many correlated factors must be considered, wildlife being one. Each of the many controlling factors must be evaluated so that in the formulation of basic control plans, those values may be considered and weighed against the whole.

I think it safe to say that no public mosquito control program is ever established unless the health or pest index justifies the appropriation and expenditure of public funds. Once organized and in operation, it is a matter of public trust to administer the program in a manner compatible with the general public's interest. The conservation of wildlife values is certainly a matter of public concern; however, it is recognized that the value the general public attaches to wildlife conservation varies considerably with the locality.

The direction and operation of a mosquito control program requires many skills sometimes concentrated in one person or spread over a number of individuals. In addition to administration, these skills include engineering, entomology, and public relations.

I think it would be generally agreed that the great majority of those directing mosquito control programs are sportsmen who would not knowingly or intentionally approve a project that would materially destroy a wildlife resource. Yet untrained in wildlife fundamentals, the problem facing the mosquito control director is just what constitutes a wildlife resource; what its value or weight is in his particular locality and what steps might be taken to maintain that value yet accomplish his objective. In many cases, the mosquito control director is confused by the levels and nature of the attacks on the program, the majority of which might be classified as destructive criticism, without the most minute offer of assistance. These stem from, in most cases, local groups of naturalists, organic gardeners and the like who are dedicated to holding the "status quo" in face of the values associated with the public good. In some few instances State Fish and Game Departments have presented a position difficult to compromise. Consequently, confusion tends to reign, tempers are ruffled and the spirit of cooperation seeks a more congenial atmosphere.

It is not unusual for the mosquito control executive to seek advice from consultants in specialty fields such as law, engineering or accounting. Therefore, in those areas where wildlife conservation is supported by a vigorous minority, State or Federal agencies should be in a position to supply competent advisors to assist in the fair appraisal of the wildlife resource and to advise in the preparation of control plans.

This necessitates the acceptance by wildlife interests that mosquito control is highly organized by State, Regional and National Associations that are constantly striving to upgrade the profession and to establish a uniformity of plan and operation based on research and sound experience. Furthermore, the wildlife interests should recognize that, as a public function, mosquito control must produce a reasonable reduction in the annoyance level or face public disfavor and extinction. In most cases,

[1] A paper from the Proceedings of the 15th Annual Meeting of the AMCA, Salt Lake City, April, 1959.

mosquito control is performed only on private lands, consequently the character and degree of control is often dependent on ownership requirements that may limit the scope of cooperation. When those private lands become public, the responsibility for mosquito control passes to the new ownership.

By the same token, mosquito control must recognize that fish and wildlife preservation are also in the public interest, and consistent with established values they must be maintained and supported. Therefore, in the consideration of every mosquito control project, the question of the fish and wildlife value attached to it must be answered.

To this end we must admit a certain ignorance. But where do we seek advice? From those we believe have unfairly attacked our program or whose minds are closed to reasonable compromise? No. As public officials we must seek our answer in the top echelon of Federal and State Conservation Services. Here is where uniform policies may be established; here facility for research and educational work is available; here should come the answers in usable form.

The stage has now been set by this Association, in cooperation with the Fish and Wildlife Service, for the formation of a National Committee representing both interests for the purpose of establishing methods of coordination, at top level, for the guidance of our workers. Through guided research and the formulation of manuals, it is hoped that a mutual appreciation of purpose will develop for the betterment of both services.

So in conclusion, we must hold that mosquito control serves a multiple public function and through years of research and investigation has developed basic standards and procedures. For its own guidance it has established State, Regional and National Associations. Mosquito control recognizes that wildlife conservation has a definite public value and wishes to conserve that value consistent with the various local requirements. Mosquito control looks to the State and Federal conservation agencies for cooperation, through research, to establish those values, to assist in the formulation of large scale plans, to assume responsibilities for uniform policies and the practice of mosquito control on their own lands.

VIRGINIA MOSQUITO CONTROL ASSN.

5709 Sellger Drive, Norfolk 2, Virginia

G. A. Treakle, Cradock, President

George C. Lyon, Ocean Park, First Vice-President

Jack E. Dent, Norfolk, Second Vice-President

Philip P. Davis, So. Norfolk, Third Vice-President

Rowland E. Dorer, Norfolk, Secretary-Treasurer

ASSOCIATION NEWS

SIXTEENTH ANNUAL MEETING

of the

AMERICAN MOSQUITO CONTROL ASSOCIATION

Boston, Massachusetts

Hotel Somerset March 27, 28, 29 and 30, 1960

Preliminary Note on the Program

Some of the subjects which will be emphasized at the Boston meeting are:

Migration and dispersion of adult mosquitoes
Effects on man and animals of insecticide residues
Practical operations problems
Eastern equine encephalomyelitis
An analysis of malaria control methods
Biological control of mosquitoes
New developments in insecticide resistance

The Program Committee will welcome suggestions. Send them to W. E. Bickley, Chairman, or D. M. Jobbins, Vice Chairman of the Program Committee, or to your Regional Director.

The host association this year is the Northeastern Mosquito Control Association, which will be holding its 6th annual meeting jointly with the AMCA's 16th. All members of both associations are urged to make plans to attend.

MOSQUITOES WANTED

We have urgent need for up to 100 grams of dry adult mosquitoes and the same quantity of mayflies.

If you have a quantity on hand, or would be able to collect these for us, please contact us at once.

C. G. BLATT & COMPANY

10930 E. 25th Street **Independence, Missouri**

DR. ROBERT MATHESON (1881–1958)

Dr. Robert Matheson, Professor of Entomology, Emeritus, at Cornell University, a pioneer worker in the field of mosquito control in this country, died on December 14, 1958 at the age of 77 in Princeton, New Jersey.

Professor Matheson was born on December 20, 1881 in West River, Nova Scotia, and came to Cornell as an undergraduate student where he was to receive the degrees of B.S.A. (1906), M.S. (1907), and Ph.D. (1911). Except for the years 1907–09 during which he served as instructor, then Professor of Entomology at the South Dakota State College and State Entomologist of that State, and the year '12–'13 when he returned to Nova Scotia to serve as Assistant Professor of Zoology and Entomology in the Agricultural College and as Provincial Entomologist, Professor Matheson spent all of his professional career at Cornell. His first appointment there was in 1907 as assistant in biology, following which he held successively the posts of instructor ('10–'12), Assistant Professor of Entomology ('12 and '14–'22), Professor of Entomology ('23–'49) and Professor Emeritus 1949 until his death.

Professor Matheson was the author of three textbooks and some 125 papers in scientific journals as well as of several experiment station bulletins. He was an authority on medical entomology. Many of his scientific papers were on this subject. He is best known perhaps among mosquito control workers far and wide for his "Handbook of the Mosquitoes of North America," which was first published in 1929 and in revised edition in 1944. This Handbood made available for the first time an authoritative guide for students, public health workers and practicing entomologists in this country who were interested in mosquito identification, biology and control. The excellent line drawings, simplified keys, lucid descriptions of species and the concise information on habits included in the book did much to lay the firm foundation on which our knowledge of mosquito control in this country now rests. Another of Professor Matheson's textbooks, "Medical Entomology," published in 1932, is recognized as authoritative, well-balanced and one of the best treatises of its kind.

Besides his scientific writings, Professor Matheson contributed to mosquito control in many ways. He served as consultant on malaria control to the Tennessee Valley Authority during the first decade of that agency's existence. His sound advice on the mosquito aspects of the program was one of the important factors in TVA's development of a highly effective malaria control (and subsequent eradication) program which was to become world famous as a pattern for work of this kind. During World War II, he served as consultant on malaria control and later on malaria eradication to the U. S. Public Health Service. Students trained under him have played major roles in the control of arthropod-borne diseases both in this country and in many other parts of the world, particularly through their participation in the activities of the Armed Forces, U. S. Public Health Service, World Health Organization, and the International Cooperation Administration.

In addition to the American Mosquito Control Association, Professor Matheson's memberships included Sigma Xi, Phi Kappa Phi, Gamma Alpha, Entomological Society of America, American Society of Tropical Medicine and Hygiene, Washington Academy of Sciences, Academy of National Sciences of Chile, and American Society of Parasitologists. He was a fellow of the American Association for the Advancement of Science and in 1949 was named to the Philadelphia Academy of Science.

Professor Matheson was highly respected as a friend and as an inspiring teacher. He took a keen personal interest in his graduate students and those of us who received through him our first introduction to vital participation in biological science recall with gratitude his ability to transmit sound scientific attitudes. His home was often the scene of social gatherings at

which he and Mrs. Matheson graciously entertained his students and their wives. One remembers with pleasure the freedom and informality of those gatherings. Robert Matheson will long hold a place of high esteem and affection in the hearts of his students, co-workers and friends.

HERBERT S. HURLBUT
ARCHIE D. HESS
GEORGE H. BRADLEY

V. S. MINNICH

EDITOR'S NOTE: Following Sam Minnich's death late last May, his daughter kindly supplied Mr. Creadon with clippings of news items about her father, taken from local papers. We could think of no more fitting tribute to him than to reprint here an excerpt from these accounts. They indicate the high esteem in which Sam was held, not only by his professional colleagues in AMCA, but also by his friends, neighbors and constituents.

From the Ormond Beach *Tribune*:

MOSQUITO CONTROL BOSS, SAM MINNICH DIES AT 62.

Mr. Minnich, 62, died of a heart attack Sunday on the Riviera County Club golf course. A native of Baraboo, Wisc., Mr. Minnich came to the Halifax Area from Chicago in 1925 and later became a retail merchant. He had worked for American Bridge, where he picked up a great deal of engineering knowledge.

When the East Volusia Anti-Mosquito Control District was formed in 1938, he started a career which won him international fame. He was put in charge of the mosquito control work in the Halifax Area.

When the district was reorganized to meet expanding needs in 1954, he became its executive director.

Mr. Minnich combined his engineering knowledge with biological advice from U. S. Public Health Service and Department of Agriculture entomologists and State scientists to develop a program of mosquito control which brought him and the area international fame.

He was the first to mount a dragline on a small barge and use it for ditching swamps.

An ardent fisherman and hunter, Mr. Minnich was careful with his use of the new chemical bug killers to make sure that he did not exterminate the fish, wildlife and friendly insect populations as well as mosquitoes.

He protected the fish which eat mosquito larvae and the birds which eat adult mosquitoes. He regarded them as allies in his work of killing mosquitoes.

As an internationally known expert in his field, he authored many technical papers on mosquito control methods and experiments and conducted numerous demonstrations for visiting delegations and experts from as far away as Indonesia and many other nations.

Mr. Minnich served in the Infantry in World War I, was chairman of the Board of Deacons of Westminster Presbyterian Church at the time of his death and was a member of the American Mosquito Control Assn. (regional director of the Southeastern Division in 1956–57–58); The Elks Club (honorary life member and past exalted ruler); the Quarterback Club, the Rotary Club and the Ellinor Village Country Club.

He is survived by his widow, Mrs. Helen Minnich; two daughters, Miss Phyllis Ann Minnich of South Daytona and Mary Kathryn (Mrs. George) Sutter of Jacksonville; and a son, Frederick Samuel Minnich.

NEWS AND NOTES

AUSTIN W. MORRILL, JR.

For an Opener, We Have a Jingle or Two Submitted by Ex-Prexy Mo Hirst, the first of which goes.

A wonderful bird is the chigger,
And oh! how she digs with her digger.
Perhaps the mosquiter,
At digging can beat 'er
But damit the skeeter is bigger.
—Anonymous

The other one is as follows:

It's not his *bite*
That spoils my night.
It's his eternal buzz-zz-z
That duzz-zz-z!

This last is by Winifred Fletcher, a woman, and we think it shows obvious sex-loyalty. It's the female mosquito that bites, *Miss Fletcher,* not the male and we hereby put you on our list to receive educational literature from the American Society for the Suppression of Calling Mosquitoes "He."

While We're Still in this Foolish Mood It Is Probably a Good Time to Tell You that Uncle Harry Has Un-Retired Again. Yup. It's like we said, and we noticed nobody stepped forward to put up any bets on how long *that* retirement would last!

Harry's new job is as Vector Control Specialist with the Coos County Health Department, and it's been three months to a day. (Harry has plans for Spain and a freighter trip after his next retirement, but that's quite a ways away.) His address is Route 2, Box 598, Coos Bay, Oregon. One of his last acts before the brief retirement was to issue a colorful and informative bulletin to Washington Staters on *Mosquito Prevention and Control.* This was patterned on the sort of bulletin so effectively utilized by the California State Bureau of Vector Control and the California Mosquito Control Association, but was more detailed in its discussions of elementary aspects, as adapted to our novice Sister State. It's extension bulletin No. 534 of the State College of Washington, Pullman, Wash.

Not to be Crept Up on, Chet Robinson of Alameda County (California) has put out another, also colorful and eye-catching, bulletin which is untitled but blazons the messages that "Mosquitoes are Unnecessary" and "Mosquito Abatement Pays Dividends." The CMCA-BVC bulletins are called, "Mosquitoes about the Home" and "Mosquitoes are Your Responsibility," incidentally, and are said to have been most helpful.

Archie Hess, Our New Prexy, Writes That He Is Now Well and Fit Again and Expects to Remain So, aside from having to ingest a few pills now and then. He had alarmed us all by turning in at the N.I.H. hospital, Division of Arthritis and Metabolic Diseases, in Bethesda for a check-up and treatment, but says that, since his condition was diagnosed at an early stage, there is no indication of permanent injury. Well, that's a big relief, Archie, and we wish for you only the chocolate and peppermint flavored pills and no more aches and pains. We had asked him to give us a run-down on his new Encephalitis Research Center at Greeley, Colorado, because we knew you'd all be interested to hear about it and here is what he had to say:

"The new encephalitis research station of the U. S. Public Health Service, Communicable Disease Center, at Greeley, Colorado, is now fully staffed and in operation. The new station represents a consolidation of activities and staff previously located at Logan, Utah, and Greeley, and will form a new 'Encephalitis Section' under the Technology Branch. The primary function of the Section is to conduct field and laboratory research on the natural history and control of the arthropod-borne viral encephalitides and related problems associated with the development and utilization of water resources.

"Dr. Archie Hess, President of A.M.C.A., is Chief of the Section, Dr. Louis C. LaMotte is Chief of the Virology Unit, and Marshall B. Rainey is Chief of the Biology and Control Unit; both of these men are members of A.M.C.A. Three field stations are operated under the Greeley headquarters: the Bakersfield, California, Field Station under the direction of Dr. R. E. Bellamy; the Wenatchee, Washington, Field Station under the direction of Virgil Miles; and the Taunton, Massachusetts, Field Station under the direction of Dr. Richard O. Hayes. All three field station directors are A.M.C.A. members.

"Dr. George Browning is Medical Epidemiologist for the Encephalitis Section, working out of Greeley headquarters. Summer field study areas are maintained in eastern Oregon; northern Utah; Grand Junction, Colorado; Fargo, North Dakota; the lower Rio Grande Valley; and the Texas Panhandle.

"In addition to the research staff, a technical assistance staff of biologists and engineers from the State Aid Section is assigned to the Greeley Station. Louis J. Ogden and Fred C. Harmston are the principal members of this staff; both are members of A.M.C.A. Among their other responsibilities is to develop joint recommendations with State Health Departments for the prevention and control of mosquitoes and other vectors associated with Federal water resource development projects.

"Any members of A.M.C.A. who happen to be in the Denver area are invited to visit the Greeley Station."

Its sounds mighty wonderful and we know that a lot of us are going to be the beneficiaries of all this in the near future. Seems as if it'll be quite an idea to add Greeley to the itinerary on that vacation trip to the Great Open Spaces, and we'll bet a lot of folks accept that invitation.

WE DON'T KNOW WHY THEY ALWAYS TALK ABOUT FLIES WHEN TELLING HOW MANY ONE PAIR COULD PRODUCE IN A SEASON BECAUSE MOSQUITOES LAY JUST ABOUT THE SAME NUMBER OF EGGS and anyone who has fought *Aedes* will attest that a high proportion of them hatch. But anyway, the Shell Oil Co's *Scan* says that a pair, if unchecked, can produce 191,010,000,000,000,000,000 between April and August, a somewhat higher figure than the August to June figure previously quoted here (cf. *Mosquito News* 18(3):260). *Scan* quotes the National Agricultural Chemicals Association as saying that this number would cover the earth to a depth of 47 feet. This seems like a good example of the dangers of everybody quoting everybody else. Any mosquito control man knows they wouldn't be covering the earth . . . they'd be up in the air buzzing around our heads!

ANOTHER KIND OF STATISTIC, HOWEVER, IS QUOTED FROM W.H.O. in the same source to the effect that 10 years ago malaria struck down 300 million people a year and caused 3 million deaths. Through the malaria eradication program, which has been, as we all know, sparked by many AMCA members, these figures have already been cut in half. Aside from the great contribution being made to mankind, the campaign is wiping out serious economic losses. In India alone, it is estimated, malaria has been causing a loss of $500 million a year in productivity. But the total cost of malaria eradication in India, anticipated by 1965, is estimated at only $114 million. You see, we are not alone.

BRUCE BROCKWAY IS LIVING UP TO EXPECTATIONS AS AN ENERGETIC MEMBERSHIP CHAIRMAN and has sent us a copy of a letter he has sent to members who had let themselves get sort of inactive, and also to the regional directors. His letter, accompanied by a self-addressed envelope, was breezy and informal and friendly, like Bruce himself, and we hear that it was pretty effective, too. It's easy to sort of get in the habit of sitting back and letting things slide, sometimes, but even AMCA, with all its fine, capable members, can't afford having any member come to the conclusion he's *dispensable*. We can all help Bruce help *us* in this.

The Skeeter OF THE VIRGINIA MOSQUITO CONTROL ASSOCIATION reports proudly that the movie, "Mosquito Control in Virginia" is now completed and available for showing. ROLLIE DORER was technical advisor and practically the entire mosquito control personnel of Tidewater Virginia served as actors and prop-men. Mr. Wade H. Williams, the distinguished narrator of the open air drama, "The Confederacy," presented each summer at Virginia Beach, is the narrator of the film, which is in full color and runs about 12 minutes. It is expected that prints will be seen by many thousands of people in the months to come and that it will play a vital role in public education and the creation of sound public support.

A PARIS GREEN LARVICIDE MADE BY COATING VERMICULITE GRANULES WITH A STICKER AND THEN WITH PARIS GREEN is also reported in *The Skeeter*, which says the operation is done with a small cement mixer which can turn out about 32 pounds in 20 minutes. The first reports, it adds, on its use in the field sound good. Meanwhile, we have heard also from others who report that the use of the vermiculite was proving a most effective carrier. Many of you no doubt have read the papers on the subject in *Pest Control* for April by AMCA'ers WILLIAM R. HORSFALL, OSCAR V. LOPP and W. J. BUCHANAN. This was the issue distributed at the Salt Lake City meetings. Anyone care to comment? We'd like some Operational Notes, you all.

LES BRUMBAUGH AND HIS EFFICIENT SAN JOAQUIN MOSQUITO ABATEMENT DISTRICT (California) were the subject for an extensive write-up in the *Stockton* (Calif.) *Record* recently. In addition to big pictures of Les at a mosquito light trap, a three-wheel motorcycle catch-basin spray unit at work and a knapsack sprayer being wielded on a ditch by an exchange student, a Mr. Yon Setyamono of Indochina, there was a full column and a half coverage of a recent tour Les led his trustees on in a "mosquito hunt." Only four years old, Les's District is a highly active one and the cooperation of the citizenry has already become so good that the few lapses are glaringly apparent. The newspaper article pinpointed some of these and offered a good example of how the public can be made aware of what it can do to promote this general welfare.

BILL BICKLEY, WHO HAS SENT US SO MUCH GOOD MATERIAL, HAS FORWARDED THE FOLLOWING REVIEW which is on a subject that is of much concern to most workers:

" 'Controlling Plant and Animal Pests in Farm Ponds with Copper Sulfate' is the title of a booklet presented by the Phelps Dodge Refining Corporation, 300 Park Avenue, New York 22, New York. This booklet is available upon request from the Phelps Dodge Corporation. There is no mention of mosquitoes. However, mosquito control workers

frequently need to eliminate algae or other aquatic plants in fresh water ponds, since many mosquito larvae are protected by some of these plants. The booklet is beautifully illustrated and gives specific information on concentrations of copper sulfate for controlling different kinds of plants. It also explains how to determine the surface acreage and the approximate volume of water in a pond. Incidentally, copper sulfate is useful in controlling leeches."

Thanks, Bill. We have often been asked for information on this subject and imagine others have too. Snails as well as leeches might have been added. These are sometimes a matter of worry because of "swimmer's itch," although fortunately not because of schistosomiasis here in North America, at least; we have even been called in to control clams being drawn into a fire-sprinkler system and growing up big enough to clog the nozzles. Mosquito controllers get into everything, sooner or later.

Bill Also Sent Us Some Information on a Luncheon Held Recently in Washington D. C. by the National Agricultural Chemicals Association, in conjunction with the Entomological Society of America, on the subject of "Research Progress in Insecticide Resistance." It was the sort of meeting that makes you wish we already had developed television in this country. Luncheon guest speaker on the first day was Tony Brown who discussed the aspects of resistance in the WHO programs with which he is so familiar. Bill McDuffie, Ken Quarterman and E. F. Knipling (whom we'll call *Knip* while we're name-dropping, because that's what we call him) all had discussions of the resistance problem, as did many other famed research workers like Dr. R. L. Metcalf, Dr. C. W. Kearns, Dr. James Crow and many others who are not AMCA'ers but whose interests nonetheless include mosquitoes and give great aid to those of us who are more deeply concerned with them. Dr. Justin Andrews, known to many of us who were in military malaria control especially, remarked on the fact that the resistance of bacteria to antibiotics might well have a relationship to our understanding of resistance in insects and pointed out the continuing need for additional work in insect physiology. Research is no longer an "ivory tower" and an impractical matter of no concern to "practical control men," as none of us needs to be reminded. It was also pointed out by one of the spokesmen for the chemical industry that currently over $20 million has been spent in research on pesticides of recently developed types.

George A. Thompson and Chet Robinson got together in their reports for the past three months by showing that you just can't please everybody. George's plaint was that they had too much rainfall; Chet's, that they had too little. George had salt marshes diluted and breeding *Culex* and influxes of adults from rice paddies. Chet had catch basins. Both had situations well in hand.

Don Grant, in Addition to His Other Duties, at the San Mateo (Calif.) Mosquito Abatement District, Is Chairman of a Research Committee Set Up by the C.M.C.A. and he has given us a small peek into the recent workings of that committee. They conducted a poll of the control men to find out in what fields they felt a need of further research. Naturally resistance is a growing and ever-present problem haunting us all, and it is not surprising that toxicology and biological control led the list. Source reduction methods, physiology and ecology followed closely, with the development of new equipment and population measurement techniques in the also-ran category. Some of the faithful laborers in the field simply hollered, "More research," apparently feeling with a certain amount of desperation that we're already doing all we *know* how to and it ain't enough. *We* think that it's like our preacher said last Sunday, most good folks tend to think they're sinners, and we think most mosquito workers in the middle of the summer feel that they aren't making half the headway they should. But the world does move, and we *are* making progress.

Glenn Collett, with Don Rees and Lewis Nielsen Looking Over His Shoulder, attests to this last fact in his Twenty-ninth Annual Report. A lot of members who had the privilege of seeing Glenn's place during the AMCA meetings this year will be interested to read that his District has secured the cooperation of the Gun Clubs in water level management and that the production of *Aedes dorsalis* and *Culex pipiens,* hitherto principal species, has been steadily reduced until they are now less than half of their former densities. Nevertheless, Glenn is not satisfied and points to the newly arisen problems of *Culex tarsalis* and *Aedes nigromaculis,* both augmented by abundant water from the mountains and deficient rainfall in the summer, causing over-irrigation, as "failures" of control. (cf. "Sinners," above.) By the vigorous action he is taking against these new menaces, we know he'll soon have *them* in retreat, too.

Fred Stutz Sent a Fine Annual Report for 1958 from His Dade County (Florida) District, which announced among many meaty statistics the sinister arrival of an anti-mosquito-fish fish. These science-fiction monsters turned out to be no product of nuclear fallout but an import from Yucatan, brought in wittingly by a fancier, like the gypsy moth, and then turned loose when he found no sale for them. Their impact on Fred's program has yet to be evaluated. He also has giant tree holes 5 to 25 feet in diameter for his tree-hole mosquitoes, with only the bright spot that the holes are so big that fish can get in and eat the mosquitoes! This brings them back to the anti-

fish fish. Fred reports his airplane spray program averaged 20 to 24 cents an acre, as against 21 for turbine ground equipment. This is quite similar to the 29 cents per acre ED WASHBURN estimated in his Turlock (Calif.) Mosquito Abatement District report for 1958, which he dropped in our lap just before taking off for Liberia, and the 20 cents their new District-owned plane has cost JOHN BRAWLEY's Kings County (Calif.) Mosquito Abatement District.

IN RESPONSE TO OUR WHO'S WHERE QUERY OF LAST ISSUE, THE CALIFORNIA BUREAU OF VECTOR CONTROL ANNOUNCES SADLY that they are beginning to wonder where everybody went, too. At the moment they have the following tally on their wall like a wartime service flag (no gold stars yet, thank goodness):

ERNIE MEYERS left for Indonesia in July; RUSS FONTAINE is still in Ethiopia (back next year, though); ED SMITH has gone back for another try at Indonesia; DON GREEN has gone to Brazil; GERRY BROOKS, has gone to Iran and thence to Ethiopia; HAL BRYDON is in Nepal; JOHN STIVERS has gone to Nicaragua; ED WASHBURN has gone to Ethiopia and ED LOOMIS has just left for Indonesia. . . .

It looks as if the next AMCA meetings might have to be in either Ethiopia or Indonesia, if this sort of migration continues.

Well, we're going to start off our Who's Who with ED LOOMIS anyway, *in absentia* or not. We had gone to all the trouble of researching him already, and picked up the phone to find his birthplace and found . . . lookie. . . . no ED!

SO WE DON'T KNOW WHERE ED LOOMIS WAS BORN but he was in California in time to go to school there. He was President of the Student Body in Junior High and High School in Oakland, giving previews of that old personality he is now so famous for. His life was largely that of athletic hero, what with varsity baseball and basketball and all, until the Navy stepped in and tapped him for two years in the South Pacific, where he rose to be an AMM 1/c and presumably learned that the world also contained mosquitoes. Back to the University of California he came to get his B.S. and then go on to graduate work and it was at this time the Bureau of Vector Control twined itself around his heart. In 1947 he became a part-time entomologist with the Bureau and in 1952 was made Associate Vector Control Specialist ("1/c," he says). For the past several years he has been Senior Vector Control Specialist at the Fresno laboratory, working on problems of measurement of mosquito populations. Ed is also a member of the American Association of Parasitologists, the Pacific Coast Entomological Society, and the Society of the Sigma Xi.

GORDON F. SMITH whom people kept mixing up with Gordon E. Smith of TVA and even with Lester Smith, at the Utah meetings, probably because they're all sort of modest and quiet, has been one of the California Mosquito Control Association's hardest working members, Vice-President in 1958 and now President in 1959. He was born in Berkeley in 1918 and received his BS in entomology from the University of California. After being with the 14th Malaria Control Detachment of the U. S. Army in the Southwest Pacific and Philippines, he was manager of the Kern County Mosquito Abatement District for ten and one-half years and then moved to the East Side MAD, at Modesto, where he is now. He is a member of the Entomological Society of America and his hobby is wood working, and he's an Optimist . . . so you see that he's a nice balance of the contemplative and studious and the practical and the gregarious.

LEWIS T. NIELSEN WAS BORN IN 1920 IN SALT LAKE CITY where he now lives and received his B.A., M.A. and Ph.D. degrees from the University of Utah, after also doing graduate work at Montana State University. He served in the Army Medical Service Corps (then known as the Sanitary Corps) from 1942 to 1945 and is still active in the Reserve, being seen from time to time on active duty as far away as the Presidio of San Francisco. He has done much fine work in mosquito taxonomy, particularly that of mountain species of *Aedes*, and also in the related fields of aquatic botany and aquatic insects. He has been a consultant for the Salt Lake City Mosquito Abatement District since 1938 and is Assistant Professor of Entomology at the University of Utah. He served as President of the Utah Mosquito Abatement Association in 1957 and 8 and has been a prime mover in the progress and activity of that very active and vigorous Association. His hobby is photography and he excels at that, too, or else he is that rare form of photo hobbyist who knows how to weed out all his bad pictures!

EDMUND J. STANSFIELD

Another loss to the ranks of mosquito control workers and the AMCA occurred with the death of Mr. Edmund J. Stansfield on February 25, 1959. He was manager of the Greater Winnipeg Mosquito Control District, a valued contributor to *Mosquito News*, and an active participant in AMCA meetings.

Good Neighbor Club

AMERICAN MOSQUITO CONTROL ASSOCIATION

5617 Sonoma Road
Bethesda 14, Maryland
August 26, 1959

Members and Sponsors of AMCA:

Due to the generosity of various sponsors, the American Mosquito Control Association was able to award Good Neighbor memberships and subscriptions to *Mosquito News* to 116 individuals in some 50 countries of the free world during 1959. The individuals selected were deserving workers who would have difficulty in obtaining U. S. currency to pay for their own membership and who would benefit from receiving *Mosquito News*. The Good Neighbor program has met with much favorable response and has done much to promote good will and exchange of information with workers in other countries. The program also has provided our commercial sponsors with an inexpensive, "non-commercial" type of advertising.

There are 150 candidates currently awaiting sponsors. Some of the candidates represent 23 countries not yet reached by sponsors through the Good Neighbor Club.

If you are not a current sponsor, we should like very much for you to participate in our 1960 Good Neighbor program by adopting some Good Neighbors. You may select the individuals or we will select them for you from the countries of your choice. Each will receive a Certificate of Award listing you as their sponsor.

Please return the tear-out form (other side of this page) with your contribution at your earliest convenience so that we can mail the Certificates to your adopted Good Neighbors before Christmas.

Thank you for supporting our Good Neighbor Program.

Sincerely yours,

Ernestine B. Thurman, Ph.D.
Chairman

MAIL TO:

Good Neighbor Club

American Mosquito Control Association
5617 Sonoma Road, Bethesda 14, Maryland

Please fill in Section A *or* B and Section C:

A. ..... We should like to sponsor the individuals whose names and addresses are given below as Good Neighbor members of AMCA for 1960.

1. .....

2. .....

3. .....

4. .....

5. .....

. .....

B. ..... We should like to sponsor ..... Good Neighbor memberships in AMCA for 1960. Please select them from the countries listed below in the order of our preference:

1. ..... 3. .....

2. ..... 4. .....

C. ..... Our check for $..... is enclosed to cover ..... memberships at $5.00 each. Please credit the sponsorship as follows on your Certificate of Award:

Name: .....

Address: .....

.....

.....
Signature

REFERENCES TO LITERATURE OF INTEREST TO MOSQUITO WORKERS AND MALARIOLOGISTS

HELEN SOLLERS
Plant Pest Control Division, Agricultural Research Service, United States Department of Agriculture

AERIAL APPLICATION

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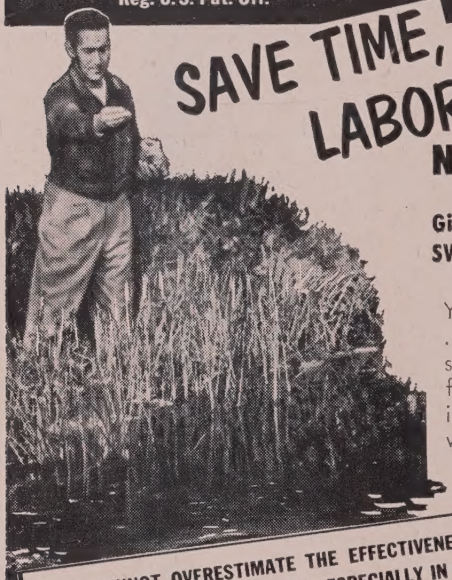

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